THE VITALITY ASSESSMENT OF TRAUMATISED PERMANENT ANTERIOR TEETH USING LASER DOPPLER FLOWMETRY

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ABBREVIATIONS

C.C. - Cardiac cycle

D.D.M. - Dichlorodifluoromethane

E.C. - Ethyl chloride

E.P.T. - Electric pulp tester

L.D.F. - Laser doppler flowmetry

P.U. - Perfusion units

S.D. - Standard deviation

S.E. - Standard error

S.W.V. - Slow wave vasomotion

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ETHICAL APPROVAL

All patients and, where appropriate, their parents gave informed consent for inclusion in this study, for which area ethical committee approval was obtained.

DECLARATION

All the work submitted herein is the original work of the author

Dafydd Evans

DEDICATION

To my father, the Rev. E. L. Parry Evans.

ABSTRACT

The clinical management of traumatised permanent anterior teeth is complicated by the unreliability of current methods of assessing dental pulp vitality. This study investigated the reliability of laser doppler flowmetry as a method of assessing the pulpal status of traumatised permanent anterior teeth.

The study found that although laser doppler flowmetry could record pulpal blood flow, non-pulpal signals were an unavoidable component of the signal obtained. The optimum non-pulpal/pulpal flux signal ratio was obtained when the laser doppler flowmeter probe was supported by a two-stage elastomeric impression jig, perpendicular to the tooth surface, at a distance of between 2-3 mm from the gingival margin. This method resulted in only 10% of the flux signal obtained from a vital dental pulp being of non-pulpal origin. Other recording methods reported in the literature were found to have between 15-45% of the flux signal of non-pulpal origin. Flux signals from vital permanent anterior teeth were found to have two characteristics which distinguished them from flux signals originating from non-vital anterior teeth; a minimum Mean Flux value of 7 perfusion units (P.U.) and a rhythmical variation in the flux signal (termed Slow Wave Vasomotion) with a frequency of between 1-10 cycles a minute, and a mean amplitude of at least 1.6 P.U.. Diagnostic criteria based on these variables were used to discriminate between flux signals from 84 vital anterior teeth and 67 non-vital anterior teeth, and were found to have a diagnostic sensitivity and specificity of 1.0.

All currently used methods of assessing dental pulp vitality were found to be less reliable (have a lower sensitivity and specificity) than laser doppler flowmetry. The sensitivity and specificity of the majority of the tests remained relatively unchanged whether applied to traumatised or untraumatised teeth. The exception were tests of

pulpal sensibility, which showed a large fall in specificity when applied to traumatised incisors as compared with untraumatised incisors. For example, it was found that vital luxated incisors were more likely than not to fail to respond positively to sensibility testing.

Although laser doppler flowmetry was found to have a sensitivity and specificity in excess of other pulpal diagnostic tests in current use, it was also found that the pulpal status of teeth was not always determined at the time of injury but could change subsequent to the injury. Both pulpal necrosis and pulpal revascularisation were possible late consequences of dental trauma, occurring months or even years following the injury. Therefore, the reliable diagnosis of pulpal vitality provided by laser doppler flowmetry should only be regarded as an aid to treatment planning the dental care of traumatised incisors. It would appear, however, that the technique of laser doppler flowmetry has potential in increasing current knowledge regarding the pathogenesis of dental pulp disease following traumatic injury.

CHAPTER 1

INTRODUCTION AND OUTLINE OF THESIS

1.1 INTRODUCTION

Dental trauma in childhood is common. Ravn (1974) reported that 35% of school-age boys and 23% of school-age girls had sustained some injury to their permanent anterior teeth. The consequences of such traumatic injury can vary from complete recovery to the loss of the tooth, trauma being the most common cause of loss of permanent anterior teeth in childhood (Todd & Dodd, 1985). Immediate loss of a tooth may occur if the tooth was so badly fractured as to be unrestorable, or if it was avulsed and not replanted. However, it is the potential for dental trauma to cause loss of vitality of the dental pulp and the consequences of pulpal necrosis, which have the greatest bearing on the long term prognosis of traumatised teeth (Andreasen & Vestergaard Pedersen, 1985). It is generally accepted that the most favourable prognosis for a tooth with an irreversibly necrotic pulp is obtained by providing appropriate endodontic therapy (Cohen, 1991). However, inadequacies of existing methods of assessing pulpal status cause difficulties in treatment planning endodontic therapy for traumatised incisors.

The dental pulp is enclosed within calcified tissue and existing methods of assessing pulpal vitality are indirect and can be unreliable. Several studies have reported a lack of correlation between clinical signs of pulpal pathology and the histopathological findings subsequent to extraction of the teeth (Seltzer, Bender & Zionitz, 1963; Tyldesley & Mumford, 1970; Moody, Browne & Robinson, 1989). In addition, existing diagnostic tests are known to be particularly unreliable when assessing the pulpal status of traumatised teeth (Magnusson & Holm, 1969; Gazelius, Olgart & Edwall, 1988) and teeth with immature root development (Klein, 1978).

A further difficulty in planning appropriate therapy for traumatised incisors is the growing awareness that under some conditions an apparently necrotic dental pulp

may revascularise and, therefore, obviate the need for endodontic therapy (Andreasen, 1986). However, the criteria on which a treatment decision to watch and wait might reasonably be made have not yet been defined. The main reason for this is that there is still no reliable method of directly assessing the pulpal status of traumatised incisors and it has recently been stated that one of the major problems facing dental traumatology is the diagnosis of pulpal and periodontal healing following traumatic injury of teeth (Andreasen, 1989).

There are indications that the new technique of laser doppler flowmetry may have considerable potential in meeting that challenge (Olgart, Gazelius & Lindh-Stromberg, 1988). Laser doppler flowmetry is an electro-optical technique which allows the non-invasive recording of blood flow through tissues and it may, therefore, allow a direct assessment of the pulpal status of traumatised teeth. At the present time, laser doppler flowmeters are expensive and the technique is time consuming. However, if laser doppler flowmetry is proved reliable, it could be used to assess the reliability of currently used methods of diagnosing pulpal status and possibly identify the clinical situations where current methods are satisfactory. It would also allow further understanding of the pathogenesis of pulpal necrosis following injury and of the pulp's potential for repair.

The hypothesis tested in this thesis is that in the management of traumatised permanent anterior teeth, laser doppler flowmetry has a diagnostic sensitivity and specificity in excess of other pulpal diagnostic methods in current use.

1.2 OUTLINE OF THESIS

An investigation into the reliability of a diagnostic method requires an understanding of the disease processes that the method is being used to diagnose. The thesis, therefore, begins in Chapter 2 with a review of the literature covering current understanding of the pathogenesis of pulpal disease following traumatic injury. Current methods of diagnosing pulpal disease are then reviewed, with laser doppler flowmetry being reviewed in context with other electro-optical methods of pulpal

diagnosis. Technical aspects of laser doppler flowmeters are further reviewed in Chapter 3.

The thesis is then divided into two main parts. The first part, covered by Chapters 3 to 6, investigates the validity of laser doppler flowmetry as a diagnostic aid for investigating pulpal disease. The relevant technical aspects of laser doppler flowmetry, including the validity and interpretation of the signal obtained from perfused tissue, are discussed in Chapter 3. Chapter 4 describes the development of the technique used in the study to record dental pulp blood flow and reviews alternative recording methods reported in other studies. Laser doppler flowmetry of the vital dental pulp is investigated in Chapter 5 and of the non-vital pulp in Chapter 6. Chapter 6 concludes with an investigation into the reliability of laser doppler flowmetry as a pulpal diagnostic aid. This section of the thesis is based on clinical data obtained from teeth which were either vital untraumatised teeth, or non-vital traumatised teeth subject to pulpectomy. The second part of the thesis, covered by Chapter 7, describes an investigation into the assessment of pulpal vitality of traumatised permanent anterior teeth using both laser doppler flowmetry and standard diagnostic methods. Chapter 8 concludes the thesis with a general review of the findings and recommendations for future research.

CHAPTER 2

LITERATURE REVIEW

2.1 INTRODUCTION

This review of the literature starts with a discussion of those aspects of the dental pulp in health and disease which are relevant to the diagnosis of pulpal pathology. Current methods of pulpal diagnosis, including the new method of laser doppler flowmetry, are then reviewed. All references to teeth in this chapter and all subsequent chapters, refer to the permanent dentition unless otherwise specified.

2.2 THE DENTAL PULP IN HEALTH

2.2.1 Introduction

The dental pulp is a highly vascular specialised connective tissue enclosed within dentine, the calcified tissue which makes up the bulk of a tooth. The primary function of the dental pulp is dentinogenesis, forming the dentine which supports the enamel covering the crown of the tooth and forming the root which supports the crown in function. Dentinogenesis commences several years before the eruption of a tooth, when cells of the primordial dental papilla are enclosed within an invagination in the enamel organ. The main process of dentinogenesis is completed around three years post-eruption when the root of the tooth is fully formed. After this dentinogenesis proceeds at a much slower rate throughout the life of the tooth. Secondary dentine is laid down on the walls of the pulp chamber, reducing its volume, until in advanced old age the pulp chamber may be virtually obliterated.

The dental pulp has other functions. It is nutritive to the dentine, which is a tubular calcified tissue. Within the proximal 1/3 of the tubules lie the odontoblastic processes, which are extensions of the odontoblasts lying within the pulp. The odontoblastic processes maintain the fluid content in the dentinal tubules and the

hydration of the dentine. The dental pulp has a defensive function: the odontoblasts responding to physical, chemical or bacterial insult by depositing calcific material within the tubules, reducing the permeability of the tissue, and by depositing reparative dentine on the walls of the pulp chamber. Pulpal tissue also contains cells of the reticulo-endothelial system such as macrophages, mast cells and lymphocytes and can mount an immune response if challenged by bacteria or bacterial by-products. The dental pulp also has a sensory function. Neural receptors are present in the dentine and the pulp, although sensations from the pulp are generally limited to those perceived as pain.

2.2.2 Pulpal haemodynamics.

The dental pulp is highly vascular: for example, in the dog the blood flow per 100 grams of tissue is the highest of all the oral tissues, though not as high as that of the heart, kidneys or spleen (Kim & Dorscher-Kim, 1989). With age, pulpal tissue becomes increasingly fibrotic, but remains perfused throughout the life of the tooth. The dental pulp receives its neurovascular supply through radicular foramina. The principal foramen is situated near to the root apex and in a mature maxillary incisor will have a diameter of about 0.4 mm, but some teeth have an additional collateral supply by way of accessory and lateral canals. Blood flow is not homogenous throughout the pulp, with coronal pulp tissue having a higher blood flow per unit of tissue than radicular pulp (Path & Meyer, 1980). The main pulpal arteries and veins (which are rather narrower than equivalent vessels in the systemic circulation) occupy the central region of the pulp. From these vessels, arterioles and venules radiate out to supply a rich plexus of capillaries in the sub-odontoblastic region (Orban, 1980). Arterio-venous shunts are present in the radicular pulp (Kim, 1990b).

2.2.3 Regulation of pulpal blood flow

The dental pulp is enclosed within rigid walls and has been described as being within a low compliance environment (Kim, 1990a). Any increase in tissue pressure cannot be rapidly equilibrated by expansion and the pulp, therefore, has some capacity for auto-regulation of blood flow. An increase in supply pressure will increase tissue pressure, increasing vascular resistance and, therefore, reduce the effect of the increase in supply pressure on blood flow. In addition, the dental pulp is innervated with afferent and efferent neurones, which may be involved in pulpal haemodynamics. Knowledge of the homeostasis of pulpal blood flow is incomplete, but the control mechanisms would appear to be complex and inter-related. Pulp circulation may be affected by stimulation of sympathetic nerves (Edwall *et al.*, 1987), sensory nerves (Gazelius & Olgart, 1980) and by perfusion with pharmacological agents (Sasano, Kuriwada & Sanjo, 1989). However, these studies were based on work on anaesthetised animals. Little is known about normal physiological control of pulpal blood flow in man, or about what factors may induce a significant change. Aars *et al.* (1992) investigated the effect of autonomic reflexes on pulp blood flow in man using laser doppler flowmetry. Three procedures known to increase sympathetic nerve activity; exercise, the cold pressor test and the isometric hand grip test were used. While all three procedures increased mean arterial pressure and heart rate, only exercise produced a consistent response, which was an increase in pulpal blood flow. However, Watson, Pitt Ford and McDonald (1992) in another study involving laser doppler flowmetry reported that exercise produced a varied pulpal response, with pulpal blood flow increasing in some subjects and decreasing in others. Aars, Brodin and Andersen (1993) investigated the effect of thermal change on human pulpal blood flow using laser doppler flowmetry and reported a variable response to both heating and cooling. They failed to find a stimulus which would provoke a reproducible acute change in pulpal blood flow.

The size of the apical foramen might affect pulpal response to stimuli. Gazelius and Olgart (1980) reported that stimulation of the cat inferior alveolar nerve resulted in an increase in wash out rate of Iodine 125 from the dental pulp. A group including Gazelius and Olgart (Edwall *et al.*, 1987), using the same technique, also reported the opposite result. The authors suggested that this was due to young cats with immature teeth being used in the first experiment, while mature cats were used in the second.

Sasano *et al.* (1989) suggested that regulation of pulpal blood flow might be more dependent on systemic arterial blood pressure than on local control. The authors investigated the effect of vasoconstrictor and vasodilator drugs on the pulpal and gingival blood flow of dogs, using laser doppler flowmetry. As anticipated, intra-venous injection of noradrenaline reduced gingival blood flow, but pulpal blood flow was found to rise along with the arterial blood pressure. Similarly, the intra-venous injection of orciprenaline, a vasodilator, increased gingival blood flow while the pulpal blood flow decreased with the fall in arterial blood pressure. This result, that gingival blood flow and pulpal blood flow in dogs are controlled by different mechanisms, is in agreement with human studies (Watson *et al.*, 1992; Aars *et al.*, 1993) and it confirmed the importance of ensuring that in pulpal blood flow studies the recording method minimises the gingival blood flow component of the measurement.

2.2.4 Pulpal neurophysiology

The dental pulp is richly innervated by myelinated (α-delta) and, more predominantly, unmyelinated (C) fibres. A pain sensory function has been shown for both types (Trowbridge, 1983). Larsson and Linde (1971) showed that some of the unmyelinated fibres are part of the sympathetic nervous system, controlling the smooth muscle of blood vessels. Variation in the sympathetic tone to the blood vessels can result in either vasoconstriction or vasodilation. Several studies have shown that the sensory innervation of a tooth may not be complete until several years post eruption. Johnsen (1985) found that unmyelinated neurones in the dental pulp reach their

maximum number soon after eruption but this may take up to five years following eruption for myelinated neurones. Fearnhead (1963) found that the parietal layer of nerves (plexus of Raschkow) develops gradually, only becoming prominent when root formation is complete. Bernick (1964), using silver staining, demonstrated a reduced number of terminal neural branches in the sub-odontoblastic layer of unerupted third molars compared with erupted controls. However, these studies were contradicted by Peckam, Torabinejard and Peckam (1991), using the more sensitive immunoperoxidase staining technique. The precise mechanism for dentine sensitivity is still uncertain. There is no doubt that dentine is innervated (Fearnhead, 1963; Byers & Matthews, 1981) but the currently accepted mechanism for dentine sensitivity is the hydrodynamic theory of Brannstrom (1963).

2.3 TRAUMA AND THE DENTAL PULP

2.3.1 Epidemiology

The national children's dental health survey of 1983 reported that 26% of all 15-year-old children had some evidence of traumatic injury to their teeth (Todd & Dodd, 1985). The report also found that trauma was the most common cause of loss of anterior teeth, resulting in the loss of 4.3 per 1000 upper central incisors in 15-year-old children. This figure will not include those teeth lost as a late consequence of their injury. The peak incidence for dental trauma is found in 9 to 10-year children (Magnusson & Holm, 1969). The anterior teeth most commonly injured following trauma are the maxillary central incisors and those least commonly injured are the maxillary and mandibular canines, comprising only 1.4% of a sample of 3,752 traumatised teeth in one study (Stalhane & Hedergard, 1975). Rock *et al.* (1974) reviewed 500 patients with a total of 801 injured incisor teeth who attended a dental hospital for emergency treatment; 88% of the affected teeth were maxillary central incisors, 7% maxillary lateral incisors and 5% were mandibular incisor teeth.

2.3.2 Pathogenesis of dental trauma

Trauma to the dento-alveolar complex may cause several types of injury. A high velocity impact, such as a road traffic accident, will tend to cause fracture of the dental hard tissues while a low velocity impact, such as an assault, will tend to displace the tooth from its socket (Oikarinen, 1987). It is unusual for a tooth to suffer both fracture and displacement (Rock & Grundy, 1981). Occasionally, a tooth may be lost as an immediate result of trauma if, for example, it is avulsed and not replanted or is so badly fractured as to be unrestorable. However, the most common complication following dental trauma is pulpal necrosis (Andreasen & Vestergaard Pederson, 1985).

Pulpal necrosis may occur in several ways. The main neurovascular supply to the dental pulp enters the root canal through the narrow apical foramen and is, therefore, vulnerable to damage if the root apex is moved. Limited movement may result in damage to the thin walled venous system resulting in venous congestion and pulpal ischaemia, while more severe displacement of the tooth may rupture the entire neurovascular supply, resulting in complete pulpal infarction. Dental trauma may also result in fracture of the crown, with the possibility of bacterial infection of the dental pulp either through direct exposure or by way of the dentinal tubules (Lin & Langeland, 1981). The precise mechanisms by which trauma can cause pulpal necrosis are still uncertain.

2.3.3 Development of pulpal necrosis

Unfortunately longitudinal studies into trauma induced pulpal necrosis do not always either classify precisely the type of injury sustained by the tooth or the stage of apical development of its root. It has been shown that these are the significant factors in determining pulpal prognosis (Andreasen & Vestergaard Pedersen, 1985). In addition, pulpal status has been assessed using diagnostic methods which are indirect and which do not necessarily reflect the true status of the pulp (Section 2.5). Also, it has been reported that the diagnosis of pulpal status may change in the weeks

following traumatic injury (Skieller, 1960; Arwill, Henschen & Sundwall-Hagland, 1967; Rock *et al.*, 1974; Rock & Grundy, 1981). The pattern of vitality was felt to be established within three months of the trauma by Skieller (1960) and Barkin (1973), within four months by Jacobsen (1980) and within six months by Zadick, Chosak and Eidelman (1979). However, this view is not supported by other authors (Magnusson & Holm, 1969; Rock & Grundy, 1981; Andreasen & Vestergaard Pedersen, 1985; Yates, 1992). These authors all reported cases where the diagnosis of pulpal status was not established until two years post-trauma.

Animal studies have limitations for the study of dental traumatology. While avulsion type injuries are easily replicated, the less serious injuries prove more problematic. For example, Tziafas (1988) reported a study on the effect of concussion injuries on the dental pulp of immature incisors. Fourteen maxillary and mandibular incisors in two dogs were subjected to experimental concussion, and examined histologicaly after 45 days. The author reported that all pulps showed marked coagulation necrosis of the coronal pulp, hyalinisation of the radicular pulp with generalised dystrophic calcifications and an absence of inflammatory infiltration. This study indicated that pulpal injury following concussion in dogs is more severe than in humans, where such an injury rarely causes pulpal necrosis (Andreasen & Vestergaard Pederson, 1985). However, the histological findings in this study were surprising in comparison with the findings of Skoglund, Hasselgren and Tronstad (1981), where extraction and replantation of incisor teeth in dogs was followed by complete revascularisation after 30 days. Instead, the high rate of pulpal necrosis reported by Tziafas for concussion injury is similar to the high rate of pulpal necrosis following intrusion injury in humans (Andreasen & Vestergaard Pedersen, 1985). Tziafas produced the concussion injury by striking the teeth in an axial direction with a metal hammer. The blow was sufficient to result in one tooth being avulsed, and another severely subluxed; both teeth being excluded from the trial. It seems likely that the lack of tooth mobility noted immediately post-trauma, which was used to classify the

injury as concussion, was in fact due to boney fixation following intrusion. In addition, the tips of the teeth were ground flat prior to injury. This would have exposed dentinal tubules, allowing bacterial ingress and would have further worsened the prognosis for pulpal revascularisation.

Despite these difficulties, careful interpretation of animal and human studies has allowed some insight into the factors influencing pulpal recovery following traumatic injury. The dental pulp is a vital tissue and if its health is compromised by physical, chemical or bacteriological injury it will attempt repair. Three factors which seem of importance in determining the outcome of pulpal repair are the processes of inflammation, bacterial infection and revascularisation and these will now be discussed.

2.3.4 Physiopathology of pulpal inflammation

The pulpal response to inflammation, which in most tissues is a healing process, is modified through the pulp being almost completely enclosed within a rigid shell (Kim, 1990a). This environment may modify the consequences of the inflammatory response such that it does not always aid pulpal healing. Kim (1990a) described pulpal inflammation as having two components; microcirculation and sensory nerve activity. The author investigated the effect of factors affecting these two components on pulpal blood flow in anaesthetised cats using laser doppler flowmetry. Three types of response were described. In Type I, pulpal blood flow was rapidly decreased by sympatheticomimetic stimuli. This would be expected, being the consequence of activation of sympathetic vasoconstrictor fibres. In the Type II response, pulpal blood flow slowly decreased following intra-arterial infusion with histamine. This was thought to be due to histamine causing an increase in vessel permeability resulting in a rise in tissue pressure and subsequent interference with pulpal circulation. A Type III response was a rise in pulpal blood flow followed rapidly by a fall, induced by intra-arterial infusion with vasodilators including Substance P, prostoglandins and bradykinins. This biphasic response was thought to be due to the vasodilation causing

passive compression of venules, raising tissue pressure and again interfering with the circulation. An alternative mechanism for circulating vasodilators to decrease pulpal blood flow was suggested by Tonder (1976), who reported that vasodilation of neighbouring vascular beds could steal perfusion pressure from the pulp. It seems, therefore, that the inflammatory response may reduce pulpal blood flow, allowing accumulation of inflammatory products and impairing pulpal healing.

Van Hassel (1971) felt the self strangulation theory of pulpal inflammation had little to commend it. The author stated that several factors would mitigate against the build up of excessive pulpal pressure, including increased lymphatic flow, increasing tissue pressure reducing the filtration pressure gradient and other capillaries having a net absorption if the tissue pressure rose above capillary pressure. Heyerass (1989) demonstrated the presence of a drainage system for fluid and macromolecules from the intercellular space in the cat dental pulp, although she did not feel able to classify it as a lymphatic system. However, Tonder and Kvinnsland (1983) have shown that these homeostatic mechanisms are insufficient to prevent a local rise in tissue pressure. In a study on anaesthetised cats the authors demonstrated that pulpal inflammation resulted in a rise in tissue pressure, from 6-10 mm Hg to 16 mm Hg. As the pulp is in a low compliance environment, the rise in pressure may interfere with circulation. This would result in a vicious circle of local accumulation of vasoactive substances, causing further vasodilation and increasing vessel permeability which in turn increases tissue pressure until pulpal circulation may be so compromised that pulpal necrosis occurs. The authors noted that the rise in tissue pressure was restricted to within 2 mm of the site of inflammation. This localisation of a rise in tissue pressure was noted by Van Hassel (1971) who pointed out that inflammation could remain localised beneath a restoration for many months without causing total pulpal ischaemia.

It may be that for inflammation to threaten the viability of the dental pulp, bacterial infection is required to provide an expanding locus of inflammatory stimulation.

2.3.5 Bacterial infection and the dental pulp

Bacterial infection is increasingly perceived as a major factor in determining the outcome of injury to the dental pulp. In the absence of bacterial infection the dental pulp demonstrates remarkable reparative potential. Gnotobiotic animal studies have shown the pulp can survive and repair following gross exposure, whether left untreated (Kakahashi, Stanley & Fitzgerald, 1965) or even if the exposure is covered by such potentially injurious agents as amalgam and zinc phosphate cement (Watts & Paterson, 1987). However, healing does not generally occur in the presence of bacterial infection (Kakahashi *et al.*, 1965; Cotton, 1974). It is presently believed that an infected pulp is irreversibly diseased (Andreasen, 1988). However, a healthy pulp can mount a successful immunological response to a bacterial challenge. Patients presenting late following traumatic exposure of a vital pulp are often found to have developed a pulp polyp, with the pulp vitality preserved. Exposure of the pulp would allow dissipation of any increase in intra-pulpal pressure caused by the inevitable pulpal inflammation. However, a vital pulp which is not exposed can also survive bacterial infection. Hoshino *et al.* (1992) demonstrated bacteria in the vital pulps of six out of nine carious molar teeth where a bridge of sound dentine remained between the carious lesion and the pulp. The survival of pulpal tissue will depend on several factors, including pulpal haemodynamics and the type and number of the infective agent. If the trauma results in vascular impairment, or even complete infarction, then it would seem likely that as an immunological response is essentially a vascular response, the ability of pulpal tissue to recover from bacterial infection would be much reduced. Bacteria may gain ingress to pulpal tissue following trauma by several routes. Firstly, traumatic exposure of a pulp chamber will allow bacteria direct access to the dental pulp. If the exposed pulp is covered with a bactericidal dressing of calcium hydroxide before the bacteria can invade deep into the coronal pulp, the prognosis for pulp survival is high (Cvek, 1978). Secondly, bacteria may penetrate dentinal tubules (Olgart, Brannstrom & Johnson, 1974; Lin & Langeland, 1981). Lundy and Stanley (1969), in an *in vivo*

study on human volunteers, demonstrated that bacteria could penetrate freshly exposed dentine. The process was not rapid, only commencing six days post exposure but after 210 days bacteria had reached a depth of 3 mm. The delay in penetration of bacteria could explain the apparent late development of pulpal necrosis in some traumatised teeth. Ravn (1981) investigated 3144 traumatised incisors with uncomplicated crown fractures and reported that the prognosis for pulp survival was linked to the amount of dentine exposed, and that early protection of exposed dentine significantly improved the prognosis. Rock *et al.* (1974) reported that treatment of a crown fracture within one month of injury significantly improved the prognosis for pulp survival.

Thirdly, bacteria may gain access to pulpal tissue by penetrating radicular dentine. Luxation type injuries will inevitably result in bacterial contamination of the root surface and in 10% of anterior teeth the cementum cover at the amelo-dentinal junction is incomplete (Armitage, 1990), facilitating bacterial penetration of dentinal tubules. Luxation injuries might also expose lateral canals. Grossman (1967), in a study involving anaesthetised dogs, found that bacteria swabbed around the gingival margins of teeth could subsequently be isolated from pulpal tissue following traumatic luxation of the teeth. The author suggested that the portal of entry was via lateral canals. Fourthly, bacteria may gain ingress to the pulp directly through the apical foramen in avulsion injuries. It is difficult to see how bacterial contamination of the pulp can be avoided when an avulsed tooth is replanted, and intriguing to speculate why overwhelming infection of the totally infarcted pulp of replanted teeth is not inevitable (Andreasen & Hjorting-Hansen, 1966). Finally, bacterial infection of a necrotic dental pulp may occur through bacteraemia, a process known as anachoresis. The role of anachoresis in pulp pathology remains controversial. If anachoresis never, or very rarely, occurs then it would be acceptable to leave traumatised teeth which are intact, but are thought to have sterile necrotic dental pulps, for very long periods to see if revascularisation might occur. Delivanis and Fan (1984) investigated the occurrence of bacterial infection of sterile necrotic canine pulps in cats. The animals were given

artificial bacteraemias at weekly intervals for two months without causing pulpal infection. Moller *et al.* (1981) also failed to find evidence of anachoresis in a study using monkeys. In both studies there was an experimentally induced total pulpal infarction. In human dental trauma this situation would tend to occur with the more severe luxation type injuries. It is possible that with concussion and subluxation type injuries the blood supply is left undamaged or only partially interrupted. It is, therefore, conceivable that if a dental pulp is only partially infarcted, the surviving circulation may allow bacteraemial infection of the damaged tissue. There is some support for this theory from the study of Delivanis and Fan (1984). As has been mentioned the authors found that sterile necrotic cat pulp tissue did not become infected, despite repeated bacteraemias. However, in two cases the periapical tissues were accidentally traumatised during an experimentally induced bacteraemia and in both cases infection of the pulp chamber contents occurred. It would seem likely that even a mild dento-alveolar injury such as subluxation will result in some damage to the periapical tissues. It is difficult to determine from human studies the significance of anachoresis in dental trauma. MacDonald, Hare and Wood (1957) reported that 83% of intact traumatised incisors, non-vital at pulpectomy, were infected. Chirnside (1957) reported that 56% of a similar sample were infected. Unfortunately, in neither study was the type of injury classified, and at least one tooth in the study by Chirnside (1957) was a replantation. Taklan (1974) reported that all 50 of a sample of intact traumatised incisors were infected. However, there might have been a problem with the method of bacterial sampling. There was no mention of a control group and the author commented on the striking similarity between the type and distribution of the organisms isolated and the oral flora. In addition, a criteria for selection into the study group was the absence of radiographic periapical pathology. With such strong evidence now linking bacterial infection to radiographic change (Section 2.8.2) it is difficult to conceive the size of the population group of non-vital incisors necessary to supply 50 teeth with infected root canals but no radiographic pathology. The weight

of evidence suggests that sterile necrotic pulps within intact teeth can become infected, although the probability of this occurring and the exact mechanism is unclear. In view of the difficulties of diagnosing pulpal status it would seem probable that loss of vitality was under diagnosed in the population groups from which the above samples were drawn. It is likely that pulpectomies were principally carried out on teeth where bacterial infection had resulted in obvious signs and symptoms, such as radiographic change, and that the incidence of sterile necrosis of pulpal tissue may not be as low as the above studies might indicate.

Loss of pulp vitality due to infarction or necrosis is not irreversible. Pulpal repair is possible through revascularisation and this process will now be discussed.

2.3.6 Pulpal revascularisation

Revascularisation of a non-perfused dental pulp may occur through anastomosis of vessels in the infarcted pulp with vessels from the periodontal ligament, through replacement of the necrotic pulpal tissue with tissue growing in from the periodontal ligament, or a combination of these two processes.

Skoglund, Tronstad and Wallenius (1978), using microangiography, noted some anastomoses occurring after only four days post-replantation of avulsed teeth in dogs. Ohman (1965) found that of eleven human premolars subject to pulpectomy between 7-14 days after replantation, three had vital pulpal tissue in contact with the roof of the pulp chamber. Ingrowth of tissue from the periodontal ligament was measured at only 0.5 mm per day, so anastomotic union of vessels would seem the likeliest explanation. In a study on pulpal enzyme activity in replanted teeth in dogs, Skoglund, Hasselgren and Tronstad (1981) found that, 10 days post-replantation, enzyme activity was generally confined to the apical root canal but some activity was noted immediately adjacent to a few large vessels in the coronal pulp. They also found that normal enzyme activity was restored throughout the pulp 30 days post-replantation. In addition, odontoblasts were noted in the revascularised pulps and further root growth

occurred. However, when the teeth were autotransplanted to a freshly prepared socket, full circulation and normal enzyme activity were not noted until 90 days post operation (Skoglund & Hasselgren, 1992); no odontoblasts were noted in the revascularised pulps and there was no evidence of further root growth. This would indicate that surviving periodontal ligament cells lining the sockets of traumatised teeth can provide progenitor odontoblast cells. If an infarcted dental pulp is revascularised, dentinogenesis will only occur if odontoblasts have survived the period of ischaemia, or if new odontoblast type cells differentiate from progenitor cells. It is known from studies on autotransplanted human premolar teeth that odontoblast function is restored in a very high proportion of cases if the transplanted tooth has an immature root (Slagsvold & Bjercke, 1974; Kristersson, 1985).

In a study on replanted premolars in humans, Ohman (1965) found that of the 44 teeth extracted more than six weeks after replantation, 83% had vital tissue in contact with the roof of the pulp chamber. Only 2% of the pulps were totally necrotic. However, it should be noted that 90% of this sample had a radiographically immature radicular apex at the time of the first extraction, a factor which significantly improves the prognosis of pulpal healing following trauma (Skieller, 1960; Andreasen, Yu & Thomsen, 1986). Mackie and Worthington (1992) reported that 15% of a sample of 34 replanted avulsed permanent incisors showed continued root growth, which indicated both pulp revascularisation and continued odontoblast function. Andreasen and Hjorting-Hansen (1966) found four out of 20 incisors with closed apices and seven out of 10 incisors with open apices showed continued root growth one year following replantation. Olgart *et al.* (1988), using laser doppler flowmetry, noted that two from a sample of four replanted permanent incisors showed restored pulpal blood flow after a few weeks. Gazelius *et al.* (1988) reported that four traumatised mandibular incisors which initially showed no blood flow using laser doppler flowmetry exhibited revascularisation of all four teeth after six weeks.

Revascularisation of a necrotic dental pulp is known to occur more often in teeth where the apical development is incomplete when assessed radiographically (Andreasen & Hjorting-Hansen, 1966; Andreasen *et al.*, 1986). However, apical development in the labial-palatal dimension lags behind development in the mesio-distal dimension and the area of the apical foramen may be larger than is indicated by a radiograph (Duell, 1973). Some authors have expressed doubt as to whether teeth with radiographically closed apices have any significant potential for revascularisation. Dumsha and Hovland (1982) followed up 52 extrusive luxations of incisor teeth with radiographically closed apices. All 52 teeth were assessed as being non-vital to electric pulp testing and pulpectomies were carried out at between four weeks and one and a half years post trauma. Only one tooth was found to have vital tissue in the coronal pulp chamber. Kling, Cvek and Mejare (1986) investigated a sample of 160 replanted permanent incisors. Of the 72 incisors with immature root apices (apex >1.1 mm) only 18% revascularised and only half of this group showed further root growth. In the other half, bone was seen to invade the pulpal lumen. None of the 88 incisors with mature apices (apex <1.1 mm) revascularised. The criteria for loss of vitality were a periapical radiolucency and/or inflammatory external root resorption. Olgart *et al.* (1988) reported on a longitudinal study of 13 luxated non-vital incisors using laser doppler flowmetry and did not note revascularisation occurring. Hasselgren, Larsson and Rundquist (1977), in a study on 13 autotransplanted human canine teeth, reported that all 13 teeth were clinically non-vital on pulpectomy, and that histochemistry failed to reveal any enzyme activity. However, it was of interest that five pulps showed diffuse calcifications as it would seem unlikely that this pathological response could occur in the complete absence of any circulation.

However, in support of the potential for revascularisation of the necrotic pulps of teeth with mature root apices, Andreasen and Hjorting-Hansen (1966) noted revascularisation in 20% of a sample of 20 replanted permanent incisors with radiographically mature apices. In addition, Gazelius *et al.* (1988) reported restored

pulpal blood flow in four luxated mandibular incisors with radiographically mature root apices. Therefore, revascularisation of teeth with mature apices can occur, although it is more likely to occur if the root apex is immature. It seems probable that it is the presence of bacterial infection which inhibits pulpal revascularisation (Kakahashi *et al.*, 1965), and it is possible that the greater surface area available for diffusion of immunological factors in immature roots improves their prognosis for revascularisation.

Teeth with necrotic dental pulps may, therefore, occasionally revascularise, although if this is going to occur the process may take some time. The consequences of pulpal necrosis for the prognosis of a tooth, and whether the prognosis is worsened if endodontic treatment is delayed until the clinical signs that the necrosis is irreversible are overwhelming, will now be discussed.

2.4 COMPLICATIONS OF PULPAL NECROSIS

2.4.1 Cessation of dentinogenesis

If the dental pulp is necrotic, odontoblast function (including dentinogenesis) will cease. The peak incidence for dental trauma in children is the ages 9 to 10-years-old (Magnusson & Holm, 1969), when the root development of the incisors may not be complete. Endodontic treatment of these teeth is possible following apexification with calcium hydroxide, but the root will remain immature, with a high pulp width/root width ratio. In addition, loss of odontoblastic function will result in dehydration of the dentine (Helfer, Melnick & Schilder, 1972) which may render it more brittle. These factors combine to weaken significantly the tooth and reduce its prognosis. Cvek (1992) reviewed a sample of 759 root treated luxated incisors four years after treatment. Of 397 incisors with immature roots, 40% had sustained a cervical fracture. Only 2% of the 362 teeth with mature root development had fractured. Pulpal healing is known to be significantly related to apical diameter, so it would seem prudent to

delay intervention in incisors with immature apical development until there are very strong clinical indications for doing so. These will be discussed in the next section.

2.4.2 Loss of pulpal defensive function

As was discussed in Section 2.3.5, pulpal tissue can mount a successful immunological response to bacterial invasion. Loss of this function with pulpal necrosis will allow bacterial infection of the root canal, which is associated with external root resorption, and the infection may spread to involve the periapical tissues. Andreasen and Hjorting Hansen (1966) reported a study of 110 replanted teeth in humans and described three types of external root resorption. There was surface root resorption, which was usually limited to the cementum, and was regarded as part of the normal homeostasis of the calcified tissues. Inflammatory external root resorption was characterised by resorption involving dentine with associated radiolucent areas, and replacement root resorption, where resorbed areas of the root were replaced with bone, with no associated radiolucency and with ankylosis. The authors reported that inflammatory root resorption was associated with pulpal necrosis and was progressive if appropriate endodontic therapy was not provided.

It has been suggested that it is the loss of viability of the cells of the cementum which results in inflammatory external root resorption (Andreasen, 1981a). However, if this is the case, it is interesting to speculate why the endodontic procedure of apicectomy, which inevitably involves loss of cementum, so rarely results in inflammatory external root resorption. Instead, it would seem to be the presence of bacterial infection of a necrotic pulp which initiates root resorption. Andreasen (1981a), in an animal study, demonstrated that infection was significantly related to inflammatory root resorption, but that non-infected necrotic pulp tissue caused only a very mild inflammatory reaction. This finding is supported by other animal studies by Makkes, Van Velzen and Van den Hooff (1978) and Skoglund and Tronstad (1981). Moller *et al.* (1981) also supported this observation in a study using monkeys, where

sterile necrotic pulp tissue left in the root canals of 26 teeth for seven months failed to provoke any radiological change. Skoglund and Hasselgren (1992) in a study on replanted teeth in dogs reported that inflammatory root resorption was associated with a necrotic pulp and that it arrested if the pulp revascularised. However, as the teeth were replanted, it seems probable that bacteria were present in the necrotic tissue.

Once such resorption is diagnosed, the treatment indicated is immediate pulpectomy and root dressing with calcium hydroxide. Inflammatory external root resorption affecting luxated teeth, with mature or immature root development, is usually successfully arrested by this procedure. Cvek (1992) reported on a sample of 197 luxated permanent incisors with external root resorption which had been treated with calcium hydroxide root dressings. At four year review, the external root resorption had been arrested in 97% of the sample. This treatment will also successfully arrest resorption affecting replanted teeth with immature roots (Andreasen, 1971), but may fail to stop resorption of replanted teeth where the root development was complete (Andreasen and Hjorting-Hansen, 1966). It has been recommended that for traumatic injuries with a high prevalence of inflammatory root resorption, such as intrusions and delayed replantation of avulsed teeth, pulpectomy should be carried out within the first two weeks of injury, without waiting for signs of pulpal necrosis (Andreasen, 1981b). However, in a review of 46 replanted avulsed incisors Mackie and Worthington (1992) found no significant relationship between the success of treatment or external root resorption when the pulpectomy was carried out electively at two weeks following injury or if it was delayed until there were clinical signs of pulpal necrosis. Inflammatory root resorption would seem to be a reliable sign of an infected necrotic pulp and an indication for immediate pulpectomy. However, with the exception of replantation of mature incisors, the prognosis for a tooth would not seem to be worsened if intervention was delayed until inflammatory root resorption had clearly developed.

Infection and bacterial products spreading from the root canal will cause inflammation in the periapical tissues and may eventually result in a dento-alveolar abscess. Acute symptomatic dento-alveolar abscess formation affecting traumatised teeth with necrotic pulps but intact crowns is a rare occurrence (Andreasen & Vestergaard Pederson, 1985). Much more common is to see radiographic evidence of periapical inflammation, the significance of which as an indicator of pulpal status will be discussed in Section 2.8.2. A retrospective study of 870 root treated traumatised incisors by Adenubi and Rule (1976) showed that existence of a periapical radiolucency at the start of treatment significantly reduced the prognosis for the treatment: the success rate falling from 91% to 82%. However, the criteria for the complete success of the endodontic treatment were very strict, being the complete absence of any radiographic periapical radiolucency. Bystrom *et al.* (1987) reported that an observation period of five years was necessary before complete healing of some pre-existing radiographic periapical lesions was noted. Similarly, Matsumoto, Nagai and Ida (1987) reported that the success rate for endodontic treatment fell from 88% to 67% if a periapical area was present before treatment. However, the follow up period was only between two and three years. In addition, the sample size was only 85 teeth and was not subdivided according to whether the root filling was adequate or was incorrectly extended. Sjogren *et al.* (1990) reviewed 635 teeth 8-10 years post endodontic treatment. The authors reported a success rate of 96% where there was no pre-existing radiographic lesion but only 86% where such a lesion was present. However, if teeth with a pre-existing radiographic lesion were adequately root treated to within 2 mm of the radiographic apex then the success rate rose to 94%, which was not significantly different from the group with no pre-existing lesion. Kerekes, Heide and Jacobsen (1980) investigated the success of endodontic therapy in 166 traumatised incisors in children. The authors reported that a pre-existing periapical lesion did not reduce the success of endodontic therapy provided that the root canal was dressed with calcium hydroxide for one year before the placement of a permanent root filling. Cvek

(1992) reported that root dressing with calcium hydroxide resulted in periapical healing in 95% of a sample of 769 luxated permanent incisors with periapical areas. Mackie *et al.* (1988) and Yates (1988) reported that pre-existing infection in non-vital teeth with open apices did not affect barrier formation time when calcium hydroxide was used to achieve apexification.

There is general agreement that endodontic treatment of a tooth with an irreversibly necrotic pulp is in the patients best interest (Cohen, 1991). If the treatment is competently carried out it will allow retention of the tooth as a functional unit within the dental arch for many years. Nevertheless, teeth which have been root treated do not have the same prognosis as teeth with vital pulps (Eckerbom, Magnusson & Martinsson, 1992). In conclusion, it would seem that delaying endodontic treatment of a non-vital tooth until there is clear clinical evidence that the pulpal necrosis is irreversible does not significantly reduce the prognosis for the tooth, provided that the endodontic treatment is of high quality and includes a pre-obturation root dressing of calcium hydroxide. The unnecessary root treatment of a vital tooth due to an error in diagnosis, or of a non-vital tooth with reversible pulpal necrosis, will reduce the prognosis for the tooth and is to be avoided.

2.5 THE CLINICAL DIAGNOSIS OF PULPAL STATUS

2.5.1 Introduction

Andreasen (1989) stated that the diagnostic shortcomings encountered in evaluating pulpal and periodontal healing subsequent to trauma were one of the major problems in dental traumatology. The fundamental problem in diagnosing pulpal status is anatomical, for the dental pulp is enclosed within a calcified shell and all existing diagnostic methods are indirect. Those methods will now be discussed.

2.6 ASSESSING PULPAL STATUS BY CLINICAL EXAMINATION

2.6.1 Symptoms of pain

Reported dental pain by the patient may result from a vital pulp becoming inflamed or from an infected necrotic pulp causing inflammation of the periapical tissues. Pain is, therefore, commonly associated with pulpal pathology caused by dental caries. Dental trauma may result in pulpal infarction, in which case classical pulpal inflammation, being a vascular phenomenon, is unlikely to occur. The necrotic dental pulp may remain sterile for prolonged periods and result in little inflammation of the periapical tissues (Moller *et al.*, 1981; Delivanis & Fan, 1984). Therefore, although pain is a useful diagnostic feature of loss of pulpal vitality caused by dental caries, it is less often a feature of loss of vitality caused by trauma (Andreasen, 1989). Dumsha and Hovland (1982) reported that in a sample of 52 luxated permanent incisors, of which 98% were subsequently proved to be non-vital by pulpectomy, only 14% of the sample were giving any symptoms. In contrast, Lin *et al.* (1984) reported that of a sample of 75 non-vital carious teeth with periapical radiolucencies, 88% were associated with pain.

2.6.2 Alveolar sinus

A discharging sinus in the alveolus is a definitive sign of infection contained within. The infection is usually associated with an infected necrotic dental pulp (Dummer, Hicks & Huws, 1980). Sinus formation is rarely seen as a sequel of loss of vitality of intact luxated incisors (Andreasen, 1989).

2.6.3 Crown colour

Coronal discolouration following trauma may occur due to extravasation of erythrocytes. Should the cells not be cleared by a functioning circulation, haemosiderin formation will result in further discolouration. The assessment of coronal discolouration is one of the more subjective diagnostic tests. Comparison with an

untraumatised antimere is not always possible and the additional test of transillumination is often not mentioned in reports in the literature. If present, coronal discolouration immediately following trauma is often described as being pinkish. Arwill, Henschen and Sundwall-Hagland (1967) described a range of 23 hues of coronal discolouration, including such subtleties as "strong violet pink plus some brown" and "grey yellow pink". Andreasen (1989) noted only grey discolouration, but there was agreement that, whatever the initial colour, there was a change to grey after about two weeks post trauma (Arwill *et al.*, 1967; Jacobsen, 1980). Discolouration may not appear for several weeks after trauma (Jacobsen, 1980) or even for 12 months or more (Yates, 1992).

There is agreement that coronal discolouration by itself is not a reliable sign of pulpal necrosis (Magnusson & Holm, 1969; Jacobsen & Sagnes, 1978; Jacobsen, 1980). However, coronal discolouration plus one other sign of pulpal necrosis (usually negative sensibility testing or radiographic change) has been regarded as a reliable indicator of pulpal necrosis (Jacobsen, 1980; Andreasen, 1986). However, there seems to be no correlation between coronal discolouration and pulpal infection (Arwill *et al.*, 1967; Andreasen, 1988), or with pulpal histology, that is whether or not the pulp is largely intact or completely autolysed (Andreasen, 1988). In addition, Andreasen (1986) noted that pulpal healing could occur even with all three signs indicating loss of pulpal vitality.

Foreman (1983) investigated the fluorescence of vital and non-vital teeth under ultra violet light. Loss of fluorescence was significantly associated with loss of vitality, and was thought to be due to quenching of the absorbed light by pulp breakdown products. However, vital and non-vital teeth could fluoresce equally strongly and the author felt the method to be too cumbersome and unreliable for routine clinical use.

2.6.4 Transillumination

Transillumination of the dental crown is an additional method of assessing colour changes in the dental tissues. If it is reported as a diagnostic test in longitudinal studies the result is usually combined with that of an examination of the labial surface of the crown, using reflected light (Jacobsen, 1980; Andreasen, 1986). Hill (1986) investigated the use of a hand held fibre optic unit in a study on 100 human permanent teeth, including both anterior and posterior teeth, which were scheduled for extraction for a variety of clinical reasons. The author reported that the method was less reliable than thermal testing with ethyl chloride. The author did comment that the test might prove to be more reliable if restricted to anterior teeth.

2.6.5 Percussion

Inflammation of the periodontal ligament may result in the patient reporting tenderness when the tooth is percussed, in comparison with the percussion of a healthy tooth. Physical injury will cause inflammation of the periodontal ligament which usually resolves within two weeks of the trauma. Teitler *et al.* (1972) reported that of 79 incisors with uncomplicated crown fractures, 36% were tender to percussion at the first assessment, which was within ten days of the trauma. However, they found no relationship between tenderness to percussion and initial or long term response to vitality testing. A positive correlation between tenderness to percussion and pulpal necrosis has been reported by Seltzer, Bender and Zionitz (1963), Garfunkel, Sela and Ulmansky (1973), Dummer, Hicks and Huws (1980) and Andreasen (1988), although Tyldsley and Mumford (1970) found no such correlation. Jacobsen (1980) reported that nearly all of a sample of 134 non-vital incisors were tender to percussion but, for reasons he did not state, dismissed the test as being of little diagnostic value. However, the author did note that the 10% of the sample which were markedly tender to percussion all had a periapical radiolucency.

Clinical signs alone, with the exception of tenderness to percussion, provide little reliable information on the pulpal status of traumatised teeth. For this reason an assessment of pulpal status includes a variety of special investigations and these methods will now be discussed.

2.7 ASSESSING PULPAL STATUS WITH TESTS OF SENSIBILITY

2.7.1 Introduction

The pulp of a tooth with completed root formation has a sensory innervation. Patients' response to stimuli applied to the crown of a tooth, such as electrical or thermal stimuli, may be used as diagnostic tests of pulpal status, although such methods only assess the sensory neural supply and, therefore, only test pulp vitality indirectly. It is accepted that although these tests are useful in the assessment of pulpal status, their results should be interpreted with caution. Several studies have demonstrated a lack of correlation between the sensory thresholds of teeth to electric pulp testing and the diagnosis following histopathological examination of the pulp (Mumford, 1967; Lundy & Stanley, 1969; Johnson, Dachi & Haley, 1970; Matthews *et al.*, 1974; Moody, Browne & Robinson, 1989). Studies have, however, shown a significant relationship between a negative response to sensibility testing and total pulpal necrosis (Seltzer *et al.*, 1963; Johnson *et al.*, 1970; Jacobsen, 1980; Moody *et al.*, 1989).

There are several reasons why sensibility testing may not always accurately reflect pulpal status. Any diagnostic test requires skill in its application and interpretation. In addition, sensibility testing relies on patients' subjective responses to the applied stimulus. This is felt to be a significant problem with regard to the tests' reliability (Ehrmann, 1977). Additional problems include the possibility that neurones may be able to function in necrotic pulp tissue, and the decreased reliability of sensibility testing in immature teeth and teeth which have been traumatised.

2.7.2 Neural tissue and pulpal necrosis.

Dental pulps which are found to be totally necrotic at pulpectomy have sometimes responded positively to sensibility testing immediately prior to treatment (Johnson, 1970; Lin *et al.*, 1984). There are three possible explanations. The first is that sensory neurones are more resistant to hypoxia than other cells. A unique feature of neural pulpal tissue, compared with other pulpal cell types, is that their cell bodies lie outside the confines of the pulp chamber. This might allow some cellular nutrition to continue even if the remaining pulpal tissue is necrotic. However, conduction of action potentials along axons requires electrolyte exchange between the axon and the supporting tissue and it seems unlikely that this system would remain unaffected by tissue necrosis. In support of this, a marked reduction in pulpal blood flow has been shown to reduce the activity of all pulpal sensory nerves (Kim, 1990a). England, Pellis and Michanowicz (1974) reported that neural tissue in human teeth did seem to have an enhanced survival capability in conditions of irreversible pulpitis compared with other cell types. However, once the tissue became necrotic neural axons, although still visible, were discontinuous and would, therefore, be unable to conduct impulses. In support of this finding Cipriano and Walton (1986) showed the myelin sheath in neural tissue from necrotic pulps to be discontinuous and concluded that the neurones were probably non-functional. Lin *et al.* (1984) reported a histopathological study on pulps extirpated from human teeth with periapical radiolucencies and noted that some teeth with necrotic dental pulps responded positively to sensibility testing. However, examination of their data shows that six teeth from the sample of 23 teeth which responded positively to sensibility testing were found to contain no pulpal tissue at all. This indicates that the technique used was capable of stimulating periodontal ligament sensory neurones. Mullaney, Howell and Petrich (1970) concluded that it was unlikely that nerve fibres were particularly resistant to necrosis.

The second explanation is that applied stimuli may activate sensory receptors in the periodontal ligament of the tooth, but are perceived by the patient as emanating

from the dental pulp. The third possibility is that sensibility testing requires a subjective response from the patient who if nervous, very young or not properly informed, may give a false positive response.

2.7.3 Pulpal sensibility in teeth with immature roots.

The delay in full sensory innervation in newly erupted teeth was discussed in Section 2.2.4. The finding that teeth with immature root development may have reduced pulpal sensibility to electric pulp testing was first reported by Reiss and Ferudi (1933). Cooke (1952) reported that an electric pulp tester sometimes failed to produce a response from teeth with radiographically immature roots. Also Mumford (1965) found that children less than 10-years-old had reduced pulpal sensibility to electric pulp testing. Fulling and Andreasen (1976a) found increasing pulpal sensibility to electric pulp testing with root development, but out of a sample of 273 anterior teeth in children only 1% failed to respond positively to electric pulp testing and all responded positively to thermal testing with carbon dioxide snow. In contrast, Klein (1978) in a study of 631 incisors in children found that for incisors where the apex was less than half closed (stages 1-5, Moorrees, Fanning & Hunt, 1963), 70% failed to respond to electric pulp testing. However, 28% of teeth with fully developed apices (stages 6-7) also failed to respond. Fuss *et al.* (1986) reported that although all 47 teeth in a sample of premolars with completed root formation responded positively to electrical stimulation, 20% of a sample of 49 premolars with incomplete root formation failed to respond. Brandt, Kortegaard and Poulsen (1988), in a longitudinal study of 34 children over two and a half years, found that pulpal sensibility to electric pulp testing increased with root development.

In conclusion the evidence would indicate that there is decreased reliability, specifically false negative results, in the sensibility testing of vital teeth with immature root formation.

2.7.4 Pulpal sensibility in traumatised teeth.

There is agreement that tests of sensibility are particularly unreliable when applied to traumatised teeth, both immediately following the trauma (Skieller, 1960; Magnusson and Holm, 1969; Teitler *et al.*, 1972; Rock *et al.*, 1974; Jacobsen, 1980; Olgart *et al.*, 1988), and for many months after (Ohman, 1965; Arwill *et al.*, 1967; Bhaskar & Rappaport, 1973; Gazelius *et al.*, 1988; Yates, 1992). It would seem likely that pulpal status following trauma may sometimes fluctuate between healing and necrosis. For example, the pulp may become non-vital immediately after a severe displacement injury due to rupture of blood vessels, or as a delayed effect due to venous congestion or late bacterial penetration of dentinal tubules (Lundy & Stanley, 1969). Similarly a necrotic pulp may repair within a few days by anastomotic union of blood vessels (Ohman, 1965; Skoglund *et al.*, 1978) or may take many weeks to regain vitality following ingrowth of tissue from the periodontal ligament (Ohman, 1965). It is, therefore, possible that a pulp which was initially necrotic following a displacement injury may revascularise, only to become necrotic again following bacterial infection. Pulpal sensibility testing which accurately indicated that sequence of events might be classified retrospectively as being unreliable, owing to the lack of direct methods of assessing pulpal status.

Sensibility tests only assess the sensory nerve supply of the dental pulp, which does not accurately reflect pulp vitality. Sensory neurones may be more susceptible to physical trauma than blood vessels and are liable to neuropraxia following pressure or stretching. Bhaskar and Rappaport (1973) reported on 25 traumatised anterior teeth which did not respond to pulpal sensibility testing, but were vital at pulpectomy. The authors suggested that as neurones tend to follow a straighter course than the more elastic blood vessels, they were more susceptible to injury by stretching. In support, Burnside, Sorenson and Buck (1974) demonstrated that teeth which were being moved orthodontically had an increased sensory threshold to electric pulp testing.

Traumatised teeth can have a functioning blood supply in the absence of sensory innervation; this was confirmed through histological examination of replanted human premolars (Ohman, 1965), through pulpectomies on traumatised incisors (Bhaskar & Rappaport, 1973) and through laser doppler flowmetry of traumatised incisors (Gazelius *et al.*, 1988). The three main methods of assessing pulpal sensibility are electric pulp testing, thermal pulp testing and the cutting of a test cavity.

2.7.5 Electric pulp testing

Almost all commercial electric pulp testers are monopolar; a single electrode is placed on the tooth and the patient completes the circuit by holding an indifferent electrode. With this design, current will pass through the entire length of the pulp canal. An alternative is the bipolar instrument, where the two electrodes are placed on opposite sides of the crown. With this design most of the current should only pass through the coronal pulp. Matthews *et al.* (1974) suggested that comparison of these two tests may distinguish between vitality in the coronal and radicular pulp. However, Robinson (1987), in a longitudinal study on 33 auto-transplanted canines and Moody, Brown and Robinson (1989) in a study on 12 non-vital human teeth, compared both tests and found no evidence that this was the case. Electric pulp testers are thought mainly to stimulate the myelinated sensory (α delta) fibres, generally not producing sufficient current to stimulate the unmyelinated fibres (Narhi *et al.*, 1979). Support for this theory comes from the gradual increase in sensibility to electric pulp testing with root maturation (Brandt *et al.*, 1988), reflecting the increase in numbers of myelinated neurones in the first years post-eruption (Johnson, 1985).

A diagnostic test may be unreliable by indicating either a false positive or a false negative result. As the dental pulp is totally enclosed, confirmation of the result

requires histological examination of the dental pulp. Studies investigating the correlation between the response to electric pulp testing and the histopathological diagnosis of pulpal status are listed in Tables 2.1 and 2.2. The results show that the main problem with the reliability of electric pulp testing in these samples were false positive results (mean % false positive rate of 11% of a sample of 380 non-vital teeth). False negative responses from vital teeth were less of a problem, occurring in only 4% of the sample of 490 vital teeth. False positive and false negative results may occur through errors in technique. The placement-site of the electrode is important; if mentioned in studies, it is often the middle third of the labial surface of the crown (Johnson, 1970; Teitler *et al.*, 1972; Matthews, 1974; Moody *et al.*, 1989; McKinstry *et al.*, 1989). Jacobson (1984) in an *in vitro* study on 31 extracted incisors demonstrated that the middle third region of the labial surface of the crown required the lowest level of current to obtain a significant current level within the pulp chamber. However, Bender *et al.* (1989) demonstrated that the lowest response threshold was obtained at the incisal edge, supporting the recommendations of Fulling and Andreasen (1976b) and Brandt *et al.* (1988). An additional reason for using the incisal edge is that it reduces the possibility of false positive responses due to stimulation of sensory receptors in the periodontal ligament. Lin *et al.* (1984) reported six teeth responding to electric pulp testing which were subsequently found to have no pulpal contents at all. Matthews (1974) built a pulp tester whose maximum current output was limited to 150 ma, 50 ma less than the stimulus threshold of periodontal sensory neurones. Unfortunately, although the instrument produced no false positive responses from teeth with totally necrotic pulps, it produced 10 false negative responses from a sample of 26 teeth with vital dental pulps.

Table 2.1 Percentage false positive response to electric pulp testing of non-vital teeth where the diagnosis was confirmed histologically.

Percentage of false positives	Number in sample	Study
28	18	Seltzer *et al.* (1963)
3	92	Mumford (1967)
43	35	Johnson (1970)
42	19	Dummer *et al.* (1980)
3	134	Jacobsen (1980)
19	31	*Lin et al.* (1984)
14	7	Moody *et al.* (1989)
Mean percentage false positives = 11% of sample of 380 teeth		

Table 2.2. Percentage false negative response to electric pulp testing of vital teeth where the diagnosis was confirmed histologically.

Percentage of false negatives	Number in sample	Study
6	18	Seltzer *et al.* (1963)
0	45	Reynolds (1966)
6	17	Mumford (1967)
1	361	Johnson (1970)
0	11	Dummer *et al.* (1980)
39	38	Moody *et al.* (1989)
Mean Percentage false negatives = 4% of sample of 490 teeth		

The incidence of false positive responses from non-vital teeth to electric pulp testing can be reduced by placing the electrode on the incisal edge of the tooth and ensuring that the tooth is dried. In support of this, Cooley, Stilley and Lubow (1984) and Bender *et al.* (1989) were able to obtain false positive results to electric pulp testing from 7% and 30% respectively of a sample of 30 root treated anterior teeth, using the gingival third of the crown. Drying the tooth and using the incisal edge reduced the false positive response rate to 0% in both studies. False positive results may also occur through a nervous patient anticipating rather than perceiving the stimulus, and in this area all such tests are dependent on the operator's communication skills for their reliability.

2.7.6 Thermal pulp testing.

The difficulty of avoiding stimulation of periodontal sensory neurones with electric pulp testing led to interest in developing reliable methods of thermal stimulation. Although it might be assumed that thermoreceptors are required to perceive thermal change, Trowbridge *et al.* (1980) demonstrated that the sensory response to thermal change occurred before there was a temperature change in the region of the pulpodentinal junction. The authors postulated that it was the hydrodynamic forces induced by the thermal change which excited the sensory nerves.

Thermal agents either lower or raise the temperature of the crown. The main agents used to decrease crown temperature are ethyl chloride, with a temperature on evaporation similar to ice from a refrigerator, carbon dioxide (-78^0 C) and dichlorodifluoromethane (DDM) (-50^0 C). Fuss *et al.* (1986) compared the reliability of these agents in a sample of 47 premolars with completed root formation, and 49 premolars with incomplete root formation. Carbon dioxide snow and DDM elicited a response from over 97% of all the teeth tested whereas ethyl chloride only elicited responses from 50% of the teeth with completed root formation and 40% of the teeth with incomplete root formation.

Unfortunately, the very low temperature of CO_2 snow produces a painful response (Fuss *et al.*, 1986) which could be a source of false positive responses. There is also some uncertainty as to whether the low temperature causes enamel infractions. Peters, Mader and Donnelly (1986), in an *in vivo* study on human enamel, reported that no damage was noted but Lutz, Mormann and Lutz (1974) and Bachmann and Lutz (1976) in similar studies using fluorescent dyes, noted some microcracks in enamel. It is possible that the relatively thick layer of calcified tissue in the crowns of premolar teeth was the cause of the poor reliability of ethyl chloride reported by Fuss *et al.* (1986). Similarly, canine teeth also have a relatively thick layer of enamel compared with incisor teeth and demonstrate a high false negative rate to ethyl chloride testing (Mumford, 1964). Robinson (1987) reported that while all of a sample of 23 healthy canines responded positively to electric pulp testing, 18% of the sample failed to respond to ethyl chloride. By contrast, all of a sample of 32 healthy incisor teeth responded positively to both ethyl chloride and electric pulp testing. Davies and Rawlinson (1988) obtained positive responses to ethyl chloride and electric pulp testing from all of a sample of 86 anterior teeth in 43 young adults.

Studies where the response to testing with ethyl chloride has been confirmed by histology are listed in Tables 2.3 and 2.4. The results show that in these samples ethyl chloride was less reliable, both in terms of false positive responses and false negative responses, than electric pulp testing. Robinson (1987) expressed concern that ethyl chloride was liable to give false positive results due to stimulation of sensory nerves within the gingival margin, periodontal ligament or the adjacent tooth. However, in a longitudinal study of 33 auto-transplanted canines he found that 25 months post transplantation all of the canines giving a positive response to ethyl chloride were also responding to electric pulp tests, making it unlikely that the ethyl chloride test was giving false positives. Testing with ethyl chloride did appear to be giving false negatives, as of all the transplanted teeth responding positively to electric pulp testing

Table 2.3 Percentage false positive response to ethyl chloride of non-vital teeth where the diagnosis was confirmed histologically.

Percentage of false positives	Number in sample	Study
22	18	Seltzer (1963)
33	18	Tyldsley *et al.* (1970)
25	8	Garfunkel *et al.* (1973)
26	19	Dummer *et al.* (1980)
18	11	Hill (1986)
29	7	Moody *et al.* (1989)
Mean Percentage false positive = 24% of sample of 81 teeth		

Table 2.4 Percentage false negative response to ethyl chloride of vital teeth where the diagnosis was confirmed histologically.

Percentage of false negatives	Number in sample	Study
5	19	Seltzer (1963)
45	11	Dummer *et al.* (1980)
10	30	Hill *et al.* (1986)
18	38	Moody *et al.* (1989)
Mean Percentage false negatives = 16% of sample of 98 teeth		

40% failed to respond to testing with ethyl chloride. However, true pulpal status was not confirmed by either histology or through continued root growth being observed on sequential radiographs.

Methods of applying heat to the surface of a tooth as a pulp vitality test include hot water (Sorensen, Phatek & Everett, 1962), heated dental hand instruments, and friction generated heat from a rubber polishing wheel (Cooley, White & Barkmeier, 1978). The method usually advocated is to use a stick of gutta percha warmed in a flame (Mumford, 1964). The degree of heating of the crown of the tooth is difficult to control and such testing can produce severe pain from an inflamed dental pulp. In addition, Zach and Cohen (1965), in an animal study, have shown that pulpal damage can occur with a rise in temperature greater than 4 degrees centigrade. Trowbridge *et al.* (1980), in an *in vitro* study on human teeth, reported that this temperature could be exceeded in the pulp chamber by the extra coronal application of hot gutta percha, although Rickoff *et al.* (1988) in an *in vivo* study on human teeth found no evidence that pulpal damage occurred. Mumford (1964) compared the reliability of heated gutta percha and electric pulp testing in the vitality testing of anterior teeth. Out of a sample of 114 teeth, all of which responded positively to electric pulp testing, only 14% gave a consistent positive response to heated gutta percha over three occasions, while 11% consistently gave a negative response. In an attempt to gain more control over the rise in temperature, Naylor (1961) developed a thermo-electrical stimulator but the device was found to be unreliable (Reynolds, 1966).

2.7.7 Test cavity

The cutting of a test cavity is felt by some authors to be the most reliable vitality test of all (Wein, 1989; Borsuk, 1990; Rowe and Pitt Ford, 1990). However, these authors were referring to the diagnosis of pulpal pathology caused by bacteria and bacterial by-products subsequent to the carious process. There is evidence that a test cavity is an unreliable indicator of pulpal vitality in traumatised teeth. Traumatic injury

may result in loss of pulpal sensibility while leaving the pulpal blood supply intact (Gazelius *et al.*, 1988). Teeth may regain sensibility following revascularisation (Robinson, 1987) but it is not certain if the dentine itself regains sensibility. The sensory innervation of the two tissues is thought to be different, with α delta fibres responsible for the sensibility of the dentine, while C fibres principally innervate the pulp itself (Narhi, 1990). An animal study by Holland, Matthews and Robinson (1987) indicated that dentine does regain sensibility following reinnervation but this is not supported by clinical experience in humans. Bhaskar and Rappaport (1973) reported on 25 cases of traumatised teeth where a test cavity resulted in a vital pulpectomy and Ohman (1965) reported that of 46 replanted human premolars which revascularised completely, none responded positively to a test cavity and in all cases the patients first response was when the pulp tissue itself was touched.

2.8 ASSESSING PULPAL STATUS RADIOGRAPHICALLY

2.8.1 Introduction

The sequelae of pulpal necrosis may cause sufficient change in the calcified tissues of the dento-alveolar complex to result in signs visible on a radiograph. For this reason radiographic examination is generally regarded as an essential part of the diagnostic process of determining the pulpal status of traumatised teeth (Magnusson & Holm, 1969; Ingle, 1976; Zadik *et al.*, 1979). It has the advantage of not requiring a subjective response from the patient, although radiographic films still require interpretation by the clinician. Diagnostic features which may be noted include periapical radiolucency, external root resorption and arrested root development.

2.8.2 Periapical radiolucency

An increase in the rate of osteoclastic activity compared with osteoblastic activity will result in the net loss of mineral from bone. It has been stated that if such lesions affect periapical bone then they must have expanded to involve cortical plate

bone before they would be visible radiographically (Bender & Seltzer, 1961). However, this study was carried out on dry mandibles. Pitt Ford (1984) demonstrated in a study in dogs, subsequently confirmed by a study on human cadavers by Lee and Messer (1986), that periapical lesions do not have to involve cortical plate bone in order to be visible radiographically. Andreasen (1986) noted that an interval of eight weeks post trauma was required for the radiographic appearance of the periodontal ligament to return to normal following a displacement injury. Therefore, any widening of the periodontal ligament space later than three months post trauma is probably pathological in origin. There is little doubt that the presence of a radiographic periapical lesion more than three months following trauma indicates pulpal disease, but it does not necessarily indicate that the disease is irreversible. Jordan, Suzuki and Skinner (1978) reported that 46% of a sample of 24 carious molar teeth with periapical radiolucencies showed complete resolution of those radiolucencies and maintained sensibility to electric pulp testing following indirect pulp capping with calcium hydroxide. Andreasen (1986) noted that luxated incisors could be affected by a transient apical breakdown (TAB) which later resolved. Pulp canal obliteration would follow in 82% of affected cases, confirming pulp survival. The finding that a periapical radiolucency may not indicate irreversible pulpal necrosis is particularly interesting in that it seems likely that bacterial infection of at least part of the pulp is required before a radiolucency will develop. Andreasen (1988) reported no relationship between the presence of bacteria in extirpated pulps and periapical radiolucency. However, in this study bacteria were demonstrated by staining rather than by bacteriological culture and this method of detecting bacteria can be unreliable (Watts, 1987).

Studies which indicate a relationship between radiographic appearance and the presence of infection within the root canal (Stabholz & Sela, 1983; Lin *et al.*, 1984; Bystrom *et al.*, 1987) generally post date studies which find no such relationship (Chirnside, 1957; MacDonald, Hare & Wood, 1957; Shovelton, 1964). It may well be that earlier studies suffered from inadequacies in the methods for detecting bacteria,

particularly the obligate anaerobes. Carlsson, Frolander and Sundquist (1977) investigated the oxygen tolerance of anaerobic bacteria from infected human pulps and found that although all 79 strains identified survived up to two hours exposure to the atmosphere, after longer periods some strains died out and after seven days only 26 strains survived. Moller *et al.* (1981) in a study using monkeys found that of a sample of 52 teeth electively devitalised and infected with oral microbes, 47 had periapical areas after seven months. Of the sample of 26 teeth where the pulp was sectioned at the level of the apical foramen, but was left *in situ* and was not inoculated with oral microbes, none of the teeth developed a periapical radiolucency. Molven, Olsen and Kerekes (1991) used scanning electron microscopy to demonstrate bacteria in the root canals of all 12 of a sample of teeth with radiographic periapical lesions.

Whether bacterial infection of the root canal can exist without causing periapical radiographic change is less clear. Taklan (1974) was able to collect a sample of 50 intact traumatised incisors with necrotic infected pulps, but with no radiographic change. However, difficulties with the validity of this study were discussed in Section 2.3.5. What is not yet known is if a periapical radiolucency indicates that a pulp is no longer perfused.

2.8.3 External root resorption

External root resorption was discussed in Section 2.4.2. It is a decisive sign of pulpal necrosis (Andreasen & Hjorting Hansen, 1966) and in a study of 134 root treated traumatised permanent incisors reported by Jacobsen (1980) affected 20% of the sample.

2.8.4 Apical root resorption

Apical root resorption is often seen in teeth following orthodontic movement, affecting 42% of maxillary permanent central incisors in one study (Cwyk, Saint-Pierre & Tronstad, 1984). Such resorption is characterised by blunting of the apex, which

retains a smooth outline on radiographic examination, a normal periodontal ligament space and the affected tooth retaining its vitality. By contrast, inflammatory apical root resorption generally causes an irregular erosion of the root apex and is associated with an increase in the periodontal ligament space. Inflammatory apical root resorption is common in teeth with apical periodontitis (Tronstad, 1988) and indicates that most, if not all, of the pulp is necrotic (Rowe & Pitt Ford, 1990). It does not, however, indicate that the pulpal necrosis is necessarily irreversible. Andreasen (1986) reported that 30% of a sample of 27 incisor teeth which regained pulpal sensibility following transient apical breakdown were affected by apical resorption.

2.8.5 Arrested root development

Evidence of continued root growth on two consecutive radiographs is a very specific sign that the dental pulp was vital for at least some, though not necessarily all, of the period separating the two films. However, continued root growth can be difficult to assess radiographicaly. Lovekin-Bennett, Smith and Orton (1988) reviewed the crown-pulp width ratio of 170 maxillary central incisors of young patients aged between 12 and 18-years-old, with a separation of two years between radiographs. The authors reported no significant change in the majority of cases. Continued apical development of an immature root can be a useful indicator of pulpal vitality, particularly if a vital antimere acts as a control. However, arrested apical development does not always indicate loss of pulpal vitality. Kling *et al.* (1986) noted arrested apical development in some of a sample of revascularised replanted teeth. Skieller (1960) also noted that traumatised anterior teeth with arrested root growth could still have pulps vital to all other criteria. Occasionally, non-vital teeth with immature roots will eventually show radiographic evidence of a calcific barrier at the root apex (Jacobsen, 1980), but this "walling off" of a necrotic pulp by the periodontium has a characteristic radiographic appearance which is not readily confused with normal apical development.

Pulp canal obliteration may occur as a pathological response to trauma (Andreasen, 1986), but the process still requires a vital pulp. Lundberg and Cvek (1980) reported that all 20 pulps extirpated from traumatised incisors affected by pulp canal obliteration were vital on histological examination.

2.9 ASSESSING PULPAL STATUS BY PHYSIOMETRIC METHODS

2.9.1 Introduction

A fundamental problem with most methods of diagnosing pulp vitality is that they do so indirectly, either through testing the function of the sensory nerve supply or by detecting signs of pulpal necrosis. As has been discussed, these methods are not always reliable and there have, therefore, been many attempts to develop methods of assessing pulp vitality more directly, by measuring parameters which would be affected by the presence or absence of a pulpal blood flow. Blood flow in the dental pulp has been investigated using several methods, including intravital microscopy (Taylor, 1950), isotope clearance (Edwall & Kindlova, 1971), hydrogen washout (Heyerass, Tonder & Aukland, 1985), and isotope labelled microspheres (Kim, 1985). However, these are invasive techniques which are not suitable for use on human subjects. Trope *et al.* (1986) reported an animal study where Xenon 133 washout was successfully used to discriminate between vital and non-vital teeth in dogs. The authors stated that, in their opinion, the technique was applicable to humans, but there have as yet been no reports of such a use.

2.9.2 Thermal Methods

Thermometric methods have been based on either demonstrating a cooler crown surface temperature on teeth with non-perfused dental pulps or demonstrating altered recovery times to a thermal challenge. For the investigation of crown surface temperature Howell, Duell and Mullaney (1970) used cholesteric crystals and Pogrel, Yen and Taylor (1989) used infra red thermography. Altered recovery times to a

thermal challenge were investigated by Fanibunda (1986), who applied heat to the crown, and Stoops and Scott (1976) who cooled the crown. All though most of these methods did allow some discrimination between vital and non-vital teeth, none were sufficiently reliable for clinical use and reports of these techniques are characterised by a lack of follow up studies.

2.9.3 Photoplethysmography and pulse oximetry

Electro-optical techniques involve transmitting light through enamel and dentine to the dental pulp and then analysing the received light for changes induced by a blood flow. There are, however, several potential problems for optical techniques. The diagnostic system will have to be highly sensitive to detect pulpal blood flow. The dental pulp occupies less than 25% of the cross sectional area of the calcified tissues of enamel and dentine within which it is encased. The calcified tissues through which the light must pass are highly mineralised and will reduce the intensity of the transmitted light due to absorption.

The diagnostic system will also have to be very specific to avoid detecting blood flow through non-pulpal oral tissues. Although enamel and dentine are not themselves perfused, teeth are situated in one of the most highly perfused environments of the body. The crystalline nature of the calcified tissues are likely to cause scattering of transmitted light, with the possibility of inadvertently detecting blood flow through the periodontium and other adjacent oral tissues. It has been reported that dentinal tubules act as optical fibres, transmitting light more apically than the position of the source on the crown surface would indicate (Walton, Outhwaite & Pashley, 1976).

Diagnostic systems based on photoplethysmography rely on detecting optical density fluctuations synchronous with the cardiac cycle. There have been promising reports of the use of these instruments for dental pulp diagnosis *in vivo* experiments on dogs (Upthegrove, Bishop & Dorman, 1966; Schmitt, Webber & Walker, 1990).

Daley *et al.* (1988), in a study on human subjects, were able to detect a pulse wave synchronous with the cardiac cycle in 70% of vital teeth tested.

Pulse oximetry utilises the different light absorption characteristics of oxygenated and de-oxygenated blood (Alexander, Teller & Gross, 1989). In this case, two wavelengths of light are transmitted through the tissues and the ratio of received light used to determine oxygen saturation. Pulse rate is detected as the highly oxygenated arterial blood at the beginning of the pulse wave replaces the de-oxygenated blood. Nissan *et al.* (1992) reported the successful use of dual wavelength spectrophotometry as a test of pulp vitality in an study using dogs. Schnettler and Wallace (1991), using a commercial pulse oximeter with a modified probe, obtained positive readings from all 44 vital incisors tested, and negative readings from all five non-vital incisors tested. However, the sample of non-vital teeth were all were endodonticaly treated. Pulse oximetry requires the transmission of light from the labial to the palatal surface of the tooth, and if the pulp chamber and root canal are filled with an opaque material, light transmission will inevitably be artificially reduced.

2.9.4 Laser doppler flowmetry

Laser doppler flowmetry is a non invasive electro-optical method for recording blood flow through tissues. The technique was first described by Riva, Ross and Benedeck (1972), when it was used to measure retinal blood flow in the rabbit. Since then there have been several hundred published reports of its use in the biological sciences, in areas ranging from circulatory physiology to plastic surgery and gastro-enterology. The physical basis of laser doppler flowmetry is described more fully in Chapter 3, but in essence light is conducted from the flowmeter to the tissue under investigation by a fibre-optic probe, which also carries efferent fibres for returning some of the reflected light back to the flowmeter. Although all the light leaving the instrument has exactly the same wavelength (due to originating from a laser source) any light reflected off a moving object (for example, red blood cells) will have an

altered wavelength, due to the doppler effect. In the instrument, photodetectors measure the interference patterns resulting from the mixing of the doppler shifted light with non-doppler shifted light, which would still have the original wavelength due to being reflected off static objects. The output from the photodetectors gives a measure of the blood flow through the tissue.

The first reported use of laser doppler flowmetry for recording dental pulp blood flow in man was by a Swedish research group (Gazelius *et al.*, 1986), with a further study and a case report from the same group in 1988 (Olgart *et al.*, 1988; Gazelius *et al.*, 1988). All three reports indicated that laser doppler flowmetry could differentiate between vital and non-vital anterior teeth in humans, with the ratio of blood flow recorded from non-vital teeth to blood flow in vital teeth averaging 0.12 across the three studies. The first study (Gazelius *et al.*, 1986) outlined the technique and reported that an infiltration injection of local anaesthetic solution containing adrenaline reduced the laser doppler flowmetry signal from a maxillary incisor by 70%. It was also found that blood flow signals from non-vital teeth had two characteristics: a reduced signal strength when compared with vital teeth and an absence of low frequency fluctuations (1-10 cycles per minute) within the signal. The case report (Gazelius *et al.*, 1988) was of restored pulpal circulation in four mandibular incisors from one patient, which initially had no detectable pulpal circulation following luxation injury. For this patient, laser doppler flowmetry diagnosed pulpal healing several months before the pulps responded to sensibility testing. The third study (Olgart *et al.*, 1988) was a longitudinal study of traumatised incisors. Twenty traumatised incisors, which were unresponsive to electric pulp testing, were followed up for a period of between 3-120 weeks, until judged vital due to positive response to sensibility testing and other clinical criteria. Sixteen of the teeth were assessed as vital to laser doppler flowmetry and went on to recover pulpal sensibility. Two teeth assessed as vital and two teeth assessed as weakly vital with laser doppler flowmetry, went on to be assessed as non-vital with laser doppler flowmetry after six to eight weeks post trauma,

with the diagnosis confirmed by pulpectomy. No non-vital teeth were reported as revascularising and no change in laser doppler flowmetry diagnosis was noted more than eight weeks following trauma. The study did indicate that electric pulp testing of traumatised teeth was an unreliable method of assessing pulp vitality, but as teeth were only included in the study if they were initially unresponsive to sensibility testing, the degree of unreliability is difficult to determine. In addition, the trauma sustained by the incisors included both subluxation and luxation injuries, so whether the reliability of sensibility testing is affected by the type of trauma sustained by the tooth cannot be determined. The placement of the electric pulp testing probe was stated as the buccal surface of the tooth, and it is possible that there was an increased risk of false positive responses from this site (Section 2.7.5). In the same study 33 teeth were diagnosed as non-vital with laser doppler flowmetry, and the diagnosis confirmed by pulpectomy. The range of values for the vital control teeth were 1.6-10 volts, and for the non-vital teeth from 0-3.0 volts. Although these values show some overlap, the laser doppler flowmetry diagnosis was based on several variables; the intra-patient ratio between the control and the non-vital tooth had a mean value of 0.13 (SE 0.03) and although low frequency oscillations and a cardiac pulse were detected in all 33 control teeth, they were absent from 88% of the 33 non-vital teeth. Finally the authors mentioned that of four replanted incisors, two had regained a pulpal blood flow as assessed by laser doppler flowmetry after a few weeks. Pettersson and Oberg (1991) reported on a hand held laser doppler flowmeter as a diagnostic aid for dental pulp vitality, but presented no data.

Laser doppler flowmetry has also been used to detect changes in dental pulp blood flow. Wilder-Smith (1988) reported significant increases in the blood flow of carious teeth in comparison to caries-free teeth, with the dressing of the carious teeth generally resulting in a return to normal blood flow values. Ramsay, Artun and Bloomquist (1991a) reported a significant decrease in pulpal blood flow in maxillary incisors following maxillary osteotomy. The studies of Watson *et al.* (1992) into

exercise, and Aars *et al.* (1992 & 1993) into the effect of exercise, heat and other factors on pulpal blood flow in man have already been described in Section 2.2.3. Christensen and Donegan (1992) presented case reports of the use of an invasive laser doppler flowmetry probe to investigate blood flow in the human masseter muscle and an inconclusive case report of the effect of pressure on pulpal blood flow in a single permanent central incisor. Ingolfsson *et al.* (1993) investigated varying the separation of the fibres in the fibre-optic probe, and found that increasing the separation increased the blood flow reading obtained from teeth. Pitt Ford, Seare and McDonald (1993) investigated the effect of local anaesthetic agents on pulpal blood flow, reporting that an infiltration injection of an adrenaline-containing local anaesthetic solution to the upper incisors produced a significant fall in pulpal blood flow. The authors did comment that although the fall in blood flow was significant it was only half that reported by Gazelius *et al.* (1986) following a similar protocol, and suggested that this might be due to a difference in the injection technique. This explanation seems unlikely and is an indication of the general assumption in many studies, with the exception of those from the Swedish group, that the signal obtained from laser doppler flowmetry of the dental pulp originates entirely from the dental pulp, and gives a linear representation of changes of blood flow through that pulp. The validity of these assumptions will be discussed in Chapter 3 and Chapter 4.

2.10 GENERAL SUMMARY

The review of the literature indicates that there is still much uncertainty over the response of the dental pulp to traumatic injury, both regarding the development of pulpal necrosis and the potential for the necrotic pulp to revascularise. A major factor impeding further progress in this field is the unreliability of current methods of assessing pulpal vitality. It is clear that laser doppler flowmetry has great potential in this area, and there is now a need to further develop the technique to determine if the method can be simplified, and to validate the criteria by which a signal can be classified as originating from a vital or non-vital dental pulp. When this has been done, the

reliability of laser doppler flowmetry as a pulpal diagnostic method can be determined and compared with other methods in current use.

CHAPTER 3

TECHNICAL ASPECTS OF LASER DOPPLER FLOWMETRY

3.1 INTRODUCTION

This chapter reviews relevant technical aspects of laser doppler flowmeters and discusses the various operating parameters for using the instruments to record dental pulp blood flow. The chapter concludes with a description of the methods used to analyse and quantify the components of the flux signal output.

3.2 THE PHYSICAL BASIS OF LASER DOPPLER FLOWMETRY

The term LASER is an acronym for Light Amplification by the Stimulated Emission of Radiation. The principle of the laser was first described by Einstein in 1917, but it was not until 1960 that Maiman constructed the first functional laser system. Due to the method of generation, laser light is monochromatic and it is this characteristic which is utilised in laser doppler flowmetry.

Laser doppler flowmetry records blood flow by detecting the doppler shift in the wavelength and, therefore, frequency of light which occurs when it is reflected from moving cells. Monochromatic light from a laser source is transmitted to the tissue under investigation by a fibre-optic cable, which also contains efferent fibres for returning some of the reflected light to the instrument. The returned light will contain a mixture of photons reflected by static objects, whose frequency will be unchanged, and photons reflected by moving blood cells whose frequency will have been shifted according to the doppler principle. The frequency of the doppler shifted light (F1) is given by the formula:

$$F = F1 \pm \frac{\frac{v}{c}}{\sqrt{1-\left(\frac{v}{c}\right)^2}}$$

where F is the frequency emitted by the source, v is the velocity of the reflector

relative to the source and c is the speed of light. Photodetectors measure the interference patterns in the returned light resulting from the mixing of the dissimilar frequencies. The signal output from the photodetectors is a function of the number of moving red blood cells which caused the doppler shift and their velocity and is referred to as the flux signal.

Flux signal	=	number of moving cells in measuring volume	X	mean velocity of moving cells

The algorithm used by the instruments signal processing unit to convert the photodetector output to the flux signal is complex and is described by Nilsson, Tenland and Oberg (1980a). However, an inherent problem with laser doppler flowmeters is that the algorithm only produces a linear relationship between the flux signal and true blood flow when the proportion of moving reflectors (blood cells) to static reflectors is low.

In order for a linear relationship to exist, photons should arrive at the photodetectors having been reflected off a single moving red blood cell and, having undergone a single doppler shift in frequency, mix with an unshifted photon. However, at high cell concentrations photons may be reflected off several moving cells before returning to the photodetectors, and will have undergone several doppler frequency shifts. A proportion of these multipli-shifted photons may, if the overall concentration of shifted photons within the returned light is high, mix with other frequency shifted photons rather than unshifted photons, resulting in a net reduction in the doppler signal. In addition, at high cell concentrations, some red blood cells will inevitably screen other red blood cells from detection by laser doppler flowmetry due to "standing in the light".

There is, therefore, a theoretical possibility that at high moving cell concentrations the flux signal from the laser doppler flowmeter will under-represent true blood flow through tissue. This hypothesis is supported by experimental

evidence from both animal and *in vitro* studies, with reports of loss of linearity when the haematocrit exceeds 1% (Vongsavan & Matthews, 1993a), 1.7% (Nilsson, Tenland & Oberg, 1980a), and 2% (Driesson *et al.*, 1990). These concentrations are usually not exceeded in the peripheral circulation of the dental pulp, but they may rise above this value in the core of the pulp (Vongsavan & Matthews, 1993b).

Comparing laser doppler flowmetry with other methods of assessing blood flow has produced some conflicting results. Kim *et al.* (1990) found a significant correlation ($p<0.004$) between laser doppler flowmetry and Xenon 133 washout in recording the decrease in pulpal blood flow in dogs following intra-venous infusion with the vasoconstrictor noradrenaline. Edwall *et al.* (1987) investigated the variations in pulpal blood flow of cats induced by several stimuli, using laser doppler flowmetry and Iodine 125 clearance. Significant correlation between the two methods was observed in recording the decrease in pulpal blood flow induced by stimulation of the cervical sympathetic trunk (correlation coefficient=0.89) and the increase in pulpal blood flow following intra-arterial infusion of the vasodilator Substance P (correlation coefficient=0.64). However, stimulation of the inferior dental nerve produced conflicting results: laser doppler flowmetry recorded an increase in pulpal blood flow while Iodine 125 clearance found it to remain unchanged, or even to decrease. The authors suggested that this was due to the two measurement techniques recording blood flow in different areas of the pulp; laser doppler flowmetry recording pulpal blood flow across most of the diameter of the pulp (a view supported by *in vitro* work by Vongsavan *et al.* (1993a)), while Iodine 125 washout reflected the blood flow through the capillaries.

In conclusion it would seem that caution should be exercised in attributing any linearity to changes in pulpal blood flow assessed using laser doppler flowmetry and that the terms "blood flow" and "flux signal" should not be regarded as interchangeable.

3.3 SIGNALS OUTPUT FROM THE LASER DOPPLER FLOWMETER.

3.3.1 Introduction

The laser doppler flowmeter used in this study was the Periflux PF2b (Perimed, Sweden), and is shown in Figure 3.1. This instrument utilises a helium/neon laser source emitting light of 632.8 nm wavelength. The fibre-optic probe used in the study contained one fibre for light transmission and two fibres for returning reflected light to the instrument, with a fibre separation of 500 microns. The probe, with the impression jig system used in the study, is shown in Figure 3.2. When recording pulpal blood flow with the PF2b flowmeter, three signal outputs can be accessed: flux, total backscatter and concentration of moving blood cells.

3.3.2 Flux signal.

The flux signal output is related to the blood flow through the tissue under investigation and this signal forms the basis of any study on blood flow using laser doppler flowmetry. A typical chart recording of the flux signal from a vital maxillary incisor is shown in Figure 3.3, where the flux signal is the red tracing between A-C. However, the flux signal is affected by the Total Backscatter signal and other operating parameters and these factors will be reviewed before a full discussion of the flux signal in Section 3.5.2.

3.3.3 Total Backscatter signal

Total Backscatter is the total signal output from the flowmeter's photodetectors, produced by both the doppler shifted and unshifted light returned by the efferent fibres in the fibre-optic probe. The Total Backscatter signal may be seen as the line D-E on the chart recording shown in Figure 3.3. The importance of the Total Backscatter signal is that it must exceed a minimum value for proper function of the signal processing unit within the flowmeter. For the PF2b this value is 2.0 volts. If Total Backscatter falls below this level, flux zero will drift and the flux signal will

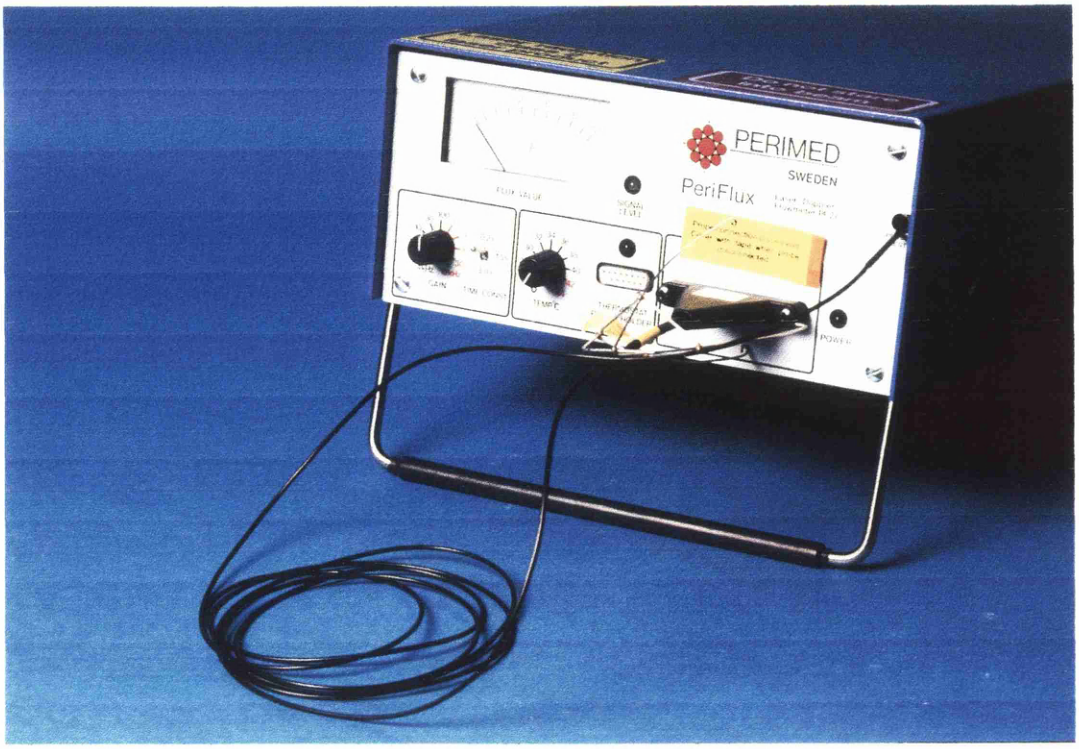


Figure 3.1 The PF2b laser doppler flowmeter (Perimed, Stockholm, Sweden).

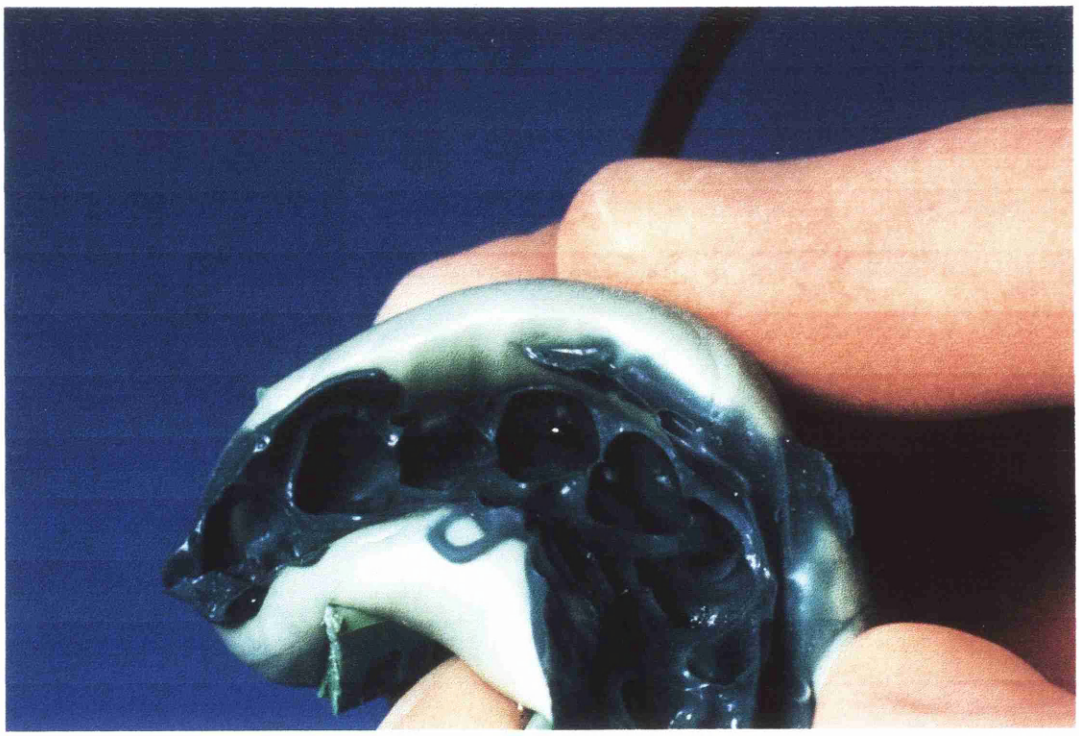


Figure 3.2 The fibre-optic probe of the laser doppler flowmeter held by a two-stage elastomeric impression jig, for recording dental pulp blood flow.

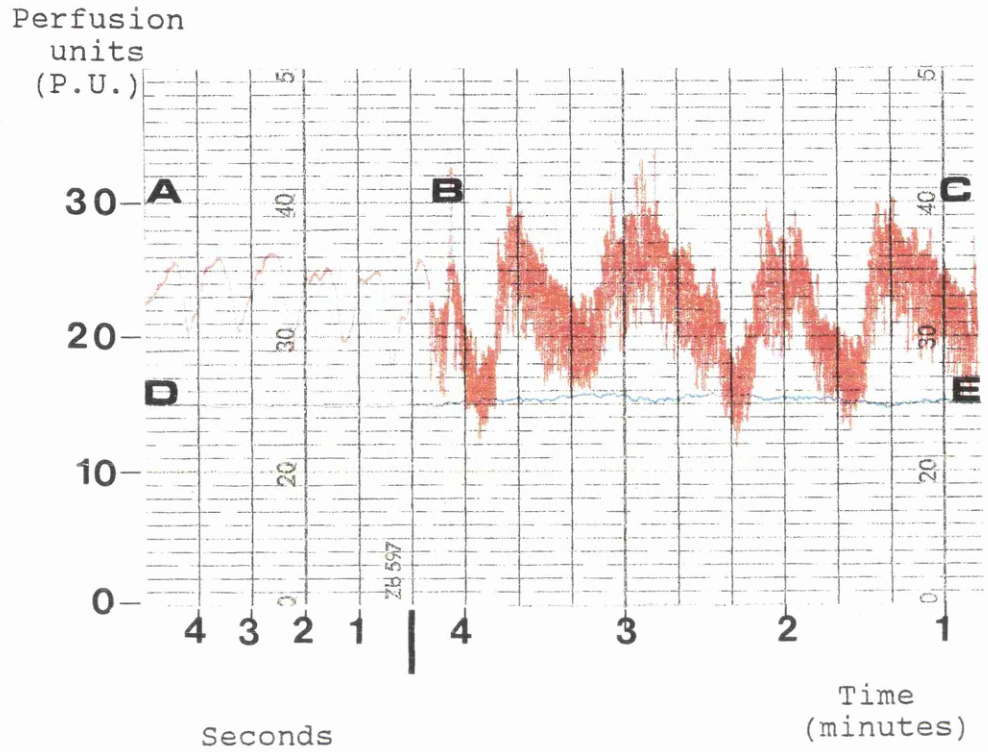


Figure 3.3 A typical flux signal from a vital maxillary central incisor.

AB- Cardiac Cycle signal, showing a mean amplitude of 5 P.U. and a regular cardiac pulse.
BC- Flux signal with a Mean Flux value of 21 P.U. and showing four cycles of Slow Wave Vasomotion, with a mean amplitude of 7.9 P.U.
DE- Total backscatter signal.

become unreliable (for discussion see Section 3.4.3). Generally, teeth give Total Backscatter values of between 2.0-3.5 volts. However, with use, the distal end of the fibre-optic probe can become scratched, resulting in reduced light transmission and difficulty in obtaining the minimum level of Total Backscatter. Application of a small drop of clear lubricating jelly to the probe tip was found to improve light transfer across the probe/enamel interface, increasing Total Backscatter by up to 1.0 volt with little effect on the flux signal.

3.3.4 Concentration of moving blood cells

This signal is proportional to the number of moving cells within the measuring volume of the laser doppler flowmeter. In view of the loss of linearity of laser doppler flowmeters at high moving cell concentrations (discussed in Section 3.2) a method of determining when the optimum cell concentration was being exceeded would aid interpretation of the flux signal. However, Vongsavan and Matthews (1993a), in an *in vitro* study on extracted teeth perfused with varying concentrations of blood, reported that the cell concentration signals of both laser doppler flowmeters tested (PF2b and Moor MBF3 (Moor Instruments, Axminster, Devon)) were very inaccurate and of little use for providing a basis for compensating for the multiple scattering of photons that occurs at high perfusion rates (Vongsavan & Matthews, 1992). This signal was, therefore, not utilised in the present study.

3.4 OPERATING PARAMETERS OF LASER DOPPLER FLOWMETERS

3.4.1 Introduction

Some aspects of the operating parameters of laser doppler flowmeters significantly affect the flux signal output. These include waveband frequency filters, zeroing the instrument and wavelength of laser source.

3.4.2 Waveband frequency filters

When a photon is reflected off a moving blood cell the frequency change due to doppler shifting will be proportional to the velocity of the blood cell; the faster the cell was moving relative to the light source, the higher the frequency shift experienced by the photon. In order to reduce signal noise, the output from the photodetectors is frequency filtered with a bandwidth filter; frequencies outside the selected band being excluded. The PF2b flowmeter has a narrow and a wide bandwidth filter. The narrow band records frequencies of between 20 Hz and 4 KHz; this frequency shift will be produced by cells with a velocity of up to 0.9 mm per second. The wide band records frequencies between 20 Hz and 12 KHz, detecting cells with a velocity of up to 2.7 mm per second (Vongsavan and Matthews, 1993b). In the young dog, the mean intravascular velocity in the primary feeding arterioles is 1.46 mm per second and in the terminal arterioles is 0.58 mm per second. In the capillaries, mean intravascular velocity is 0.27 mm per second and in the venules is 0.57 mm per second (Kim & Dorscher-Kim, 1989). The narrow 4 KHz waveband filter will, therefore, mainly record blood flow in the terminal arterioles, capillaries and venous system. These blood vessels are concentrated in the peripheral pulp, with the larger vessels with their higher blood flow velocities being more centrally placed (Orban, 1980).

Vongsavan and Matthews (1993a) advised that the wide 12 KHz bandwidth filter, which would allow detection of the flow through the large central vessels, should be used in order to reproduce as accurately as possible the total blood flow throughout the pulp. However, in their *in vitro* study the authors reported that the linearity of flowmeters was unreliable at erythrocyte concentrations in excess of 1% v/v, a concentration which is exceeded in the central vessels (Vongsavan & Matthews, 1992). There would, therefore, seem to be some advantage to the linearity of the blood flow recordings through having the greater proportion of the signal originating from peripheral vessels.

Ramsay, Artun and Bloomquist (1991b) opted for the 12 KHz wide bandwidth filter when using the PF2b to investigate changes in pulpal blood flow following osteotomies. However, the authors acknowledged problems with both signal to noise ratio and drifting of the zero with this filter and stated that the narrow bandwidth filter would be used in future. Gazelius *et al.* (1986), Wilder-Smith (1988) and Olgart *et al.* (1988) all used a PF2b, and all opted for the 4 KHz bandwidth filter.

3.4.3 Zeroing the flux signal

Laser doppler flowmeters are zeroed by recording from a static reflector, ensuring that the reflected light does not contain doppler shifted frequencies. In the present study a zero value for the flux signal was obtained by placing a pellet of impression material (Provil, Bayer) over the end of the fibre-optic probe. The flux signal obtained using this method, and the flux signal recorded from the incisal tip of a non-vital tooth using the recording method used throughout this study (see Appendix A) are shown in Figure 3.4. For the flux zero to remain stable, the Total Backscatter value must not fall below 2.0 volts (Section 3.3.3). The effect of varying Total Backscatter on the flux values obtained from a static reflector using the 12 KHz and the 4 KHz waveband filters are shown in Figure 3.5. For comparison, the effect of varying Total Backscatter on the flux signal from vital teeth using the 12 KHz waveband is shown in Figure 3.6, and using the 4 KHz waveband filter in Figure 3.7. As can be seen from these figures, with the 4 KHz narrow bandwidth filter, variations in Total Backscatter above 2.0 volts had little apparent effect on either the zero flux value obtained with a static reflector or on the flux value obtained from a vital dental pulp. There would, therefore, appear to be no necessity to apply a correction factor to every flux signal depending on the Total Backscatter value at which it was obtained, as recommended by Vongsavan and Matthews (1993b). However, with the 12 KHz wide bandwidth filter, variation of Total Backscatter above 2.0 volts had an affect on the zero flux value, particularly within the range 2.0-3.5 Volts. This is the

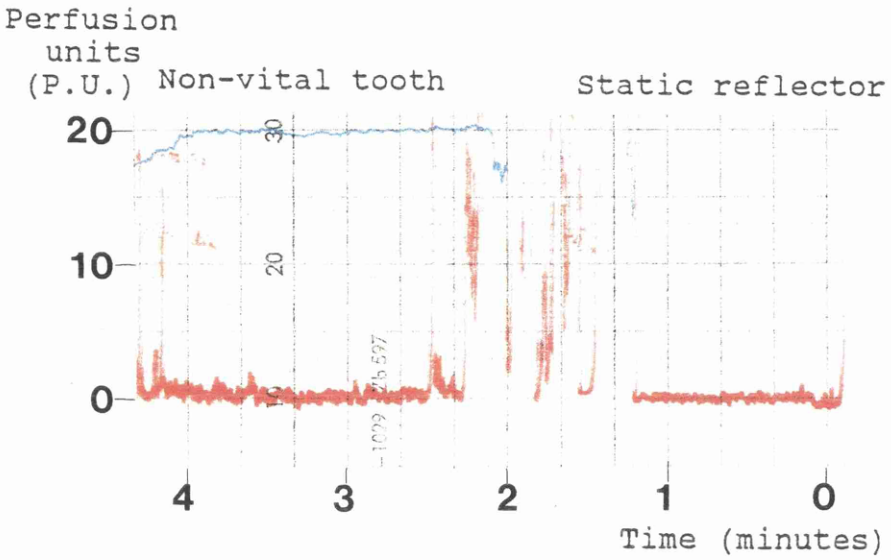


Figure 3.4 Flux signal recorded extra-orally from a static reflector (elastomeric impression material), and intra-orally from the incisal edge of a non-vital tooth, using a two stage elastomeric impression jig.

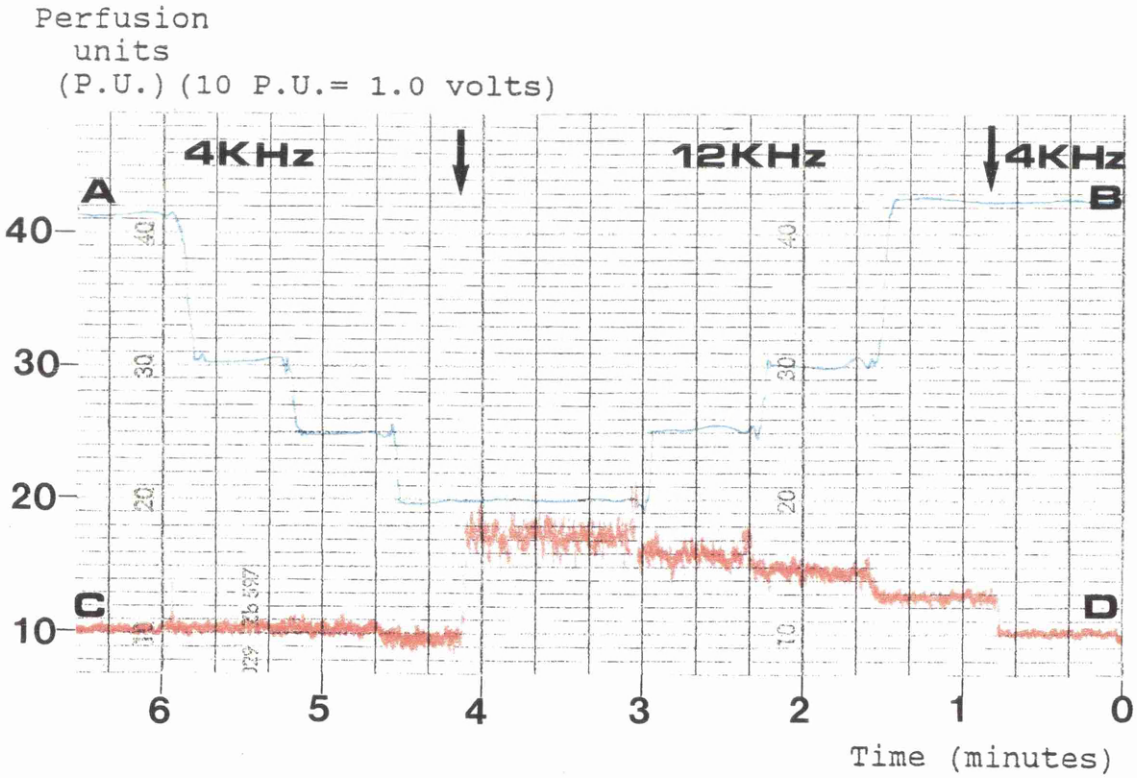


Figure 3.5 Effect of variation in Total Backscatter signal on flux signal recorded extra-orally from a static reflector, using the 4KHz waveband filter and the 12 KHz waveband filter.

AB- Total Backscatter
CD- Flux signal (zero adjusted to 10 P.U. for this recording)

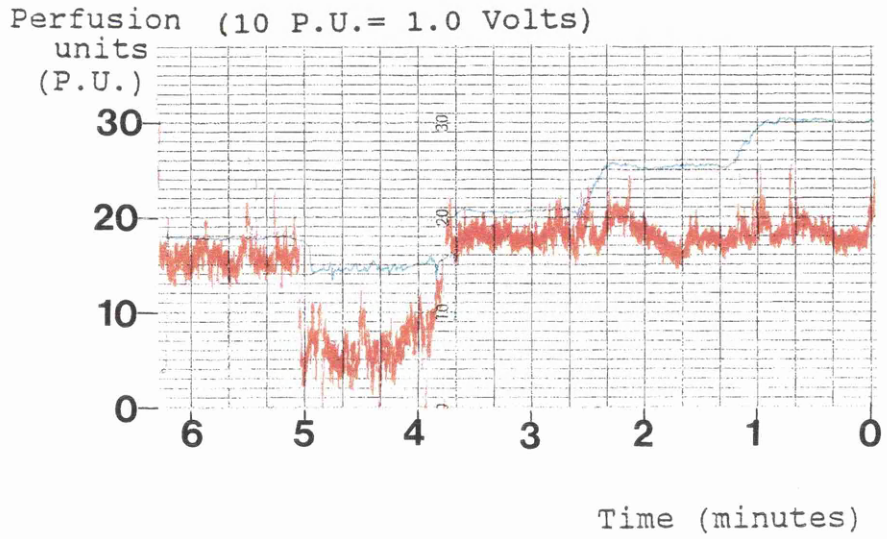


Figure 3.6 The effect of alteration of Total Backscatter on the flux signal obtained from a vital maxillary incisor, using the 12 KHz waveband filter.

Blue tracing- Total Backscatter
Red tracing - Flux signal

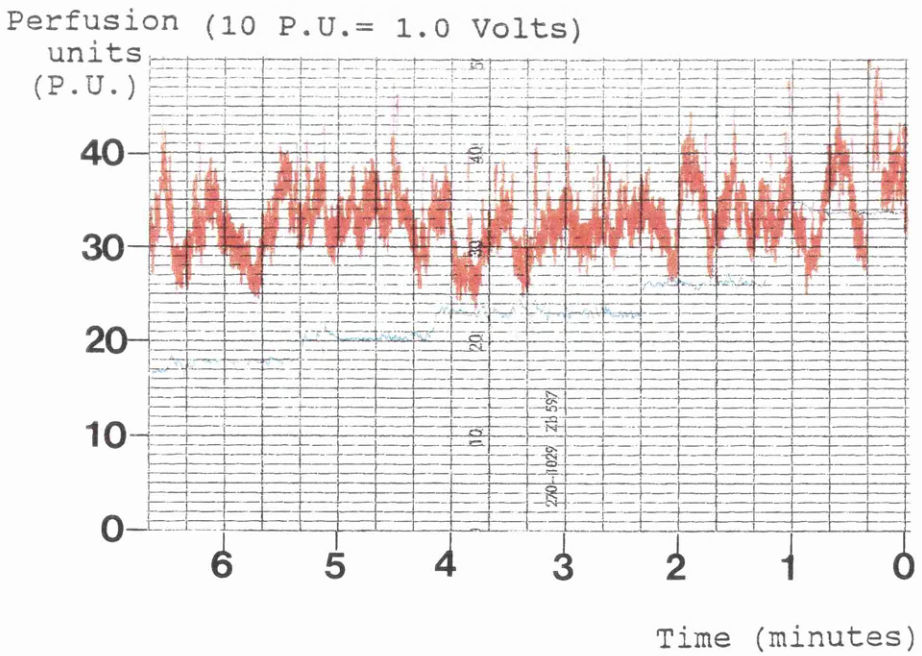


Figure 3.7 The effect of alteration of Total Backscatter on the flux signal obtained from a vital maxillary incisor, using the 4 KHz waveband filter.

Blue tracing- Total Backscatter
Red tracing - Flux signal

range of total backscatter values obtained from the majority of teeth. There would, therefore, appear to be some basis for the concern expressed by Ramsay *et al.* (1991b) as to the validity of their findings when using the wide bandwidth filter. However, the effect of variation in total backscatter on flux signals when using the 12 KHz bandwidth filter seems to be reduced when the flux output contains a high proportion of doppler shifted frequencies (Figure 3.6).

3.4.4 Wavelength of laser source

It is known that the longer the wavelength of light, the greater is its depth of penetration through a media. For example, in human skin, the depth at which light retains 37% of its incident energy is 1200 microns for light of wavelength 800 nm, but is only 550 microns for light of wavelength 600 nm (Anderson & Parrish, 1981). Petterson and Oberg (1991) demonstrated improved optical transmission through tooth tissue for a 750 nm laser source compared with a 633 nm source. The authors also claimed a reduced non-pulpal/pulpal signal ratio for the longer wavelength but, unfortunately, no data was presented to support this claim. Obeid, Dougherty and Pettinger (1990) in a well designed animal study demonstrated that under standardised conditions laser doppler flowmetry systems based on 780 nm laser sources sampled larger volumes of tissue than systems based on 633 nm laser sources. Vongsavan and Matthews (1993a) reported that in an *in vitro* study on extracted teeth under standardised conditions, larger signals were obtained from the same blood flow with a 810 nm laser source compared with a 633 nm laser source. The authors also reported that the 810 nm source sampled a greater proportion of blood flow in the core and distal periphery of the pulp chamber compared with the peripheral blood flow closest to the probe and recommended 810 nm rather than 633 nm as a laser source for laser doppler flowmetry of the dental pulp. However, a potential problem in assessing pulpal blood flow using laser doppler flowmetry is unknowingly including gingival blood flow in the measuring volume of the flowmeter (Chapter 4). The results of a

pilot study comparing laser doppler flowmeters of different wavelengths are presented in Appendix B. There were indications from this study that flowmeters with wavelengths longer than 633 nm include an increased proportion of gingival blood flow in their pulpal flux signal. Therefore, attempting to increase the pulpal flux signal by using longer wavelengths of light may be counter-productive. It is a surprising omission that none of the studies on pulpal blood flow using flowmeters with wavelengths longer than 633 nm (see Section 4.1) have reported the non-pulpal/pulpal signal ratio obtained in their study. Without this data there is no evidence that the flux signal reported originates from the dental pulp.

3.4.5 Conclusion

In conclusion there is evidence that the PF2b, with its laser source of 633 nm, records the majority of its signal from the peripheral pulp closest to the fibre-optic probe (Vongsavan & Matthews, 1993a). It is from this part of the pulp, due to the low moving cell concentration (Vongsavan & Matthews, 1992) and low cell velocities (Kim & Dorscher-Kim, 1989), that laser doppler flowmeters are most likely to give linear representation of blood flow (Obeid *et al.*, 1990). For this reason, as well as the relative stability of flux zero at 4 KHz (Section 3.4.3) the 4 KHz waveband filter was used throughout this study.

3.5 COMPONENTS OF THE FLUX SIGNAL

3.5.1 Introduction

The flux signal output supplies the data on which laser doppler flowmetry studies are based. A typical chart recording of the flux signal from a vital dental pulp is shown in Figure 3.3, and visual analysis of the chart indicates several identifiable variables which will now be discussed.

3.5.2 Mean Flux

Mean Flux is the average flux signal over the period of the recording. It is shown between B-C in Figure 3.3. Mean Flux cannot be quantified in absolute units of blood flow per unit volume as the measuring volume of tissue 'read' by laser doppler flowmeters will depend on the tissues' optical properties. In skin, the measuring volume of the PF2b flowmeter is a hemisphere with a radius of around 1 mm from the end of the probe (Nilsson *et al.*, 1980b). With teeth the measuring volume is in excess of this, and blood flow in other oral tissues can be recorded through the full thickness of the crown of a tooth. For example, Figure 3.8 shows the nearly 100% increase in flux signal recorded from the labial surface of a vital maxillary incisor when the patient placed his/her tongue on the palatal aspect of the crown during the recording. In addition, if the fibre-optic probe is angled towards the gingival margin of a tooth, the Mean Flux signal will increase, probably due to periodontal tissue on the palatal aspect of the tooth being included within the measuring volume of the flowmeter (Figure 3.9). Therefore, flux values are expressed in arbitrary perfusion units (P.U.). For the PF2b, 1 volt of flux signal output is equivalent to 10 P.U.. The gain of the instrument is calibrated monthly against a motility standard (consisting of an aqueous suspension of latex microspheres) supplied by the manufacturer. In the present study, Mean Flux was assessed visually from the chart recording. To determine the measurement error of this method, 50 charts were reassessed after an interval of one month. The results are shown in Table 3.1.

3.5.3 Slow Wave Vasomotion

Slow Wave Vasomotion describes the cyclical change in the flux signal, with a frequency of between 1-10 cycles per minute, which can be seen in Figure 3.3 between B-C. This fluctuation in flux is seen in most tissues of the body, but its function and precise physiological origin are unknown (Salerud *et al.*, 1983). Slow Wave Vasomotion does not always remain regular as can be seen in the flux signal

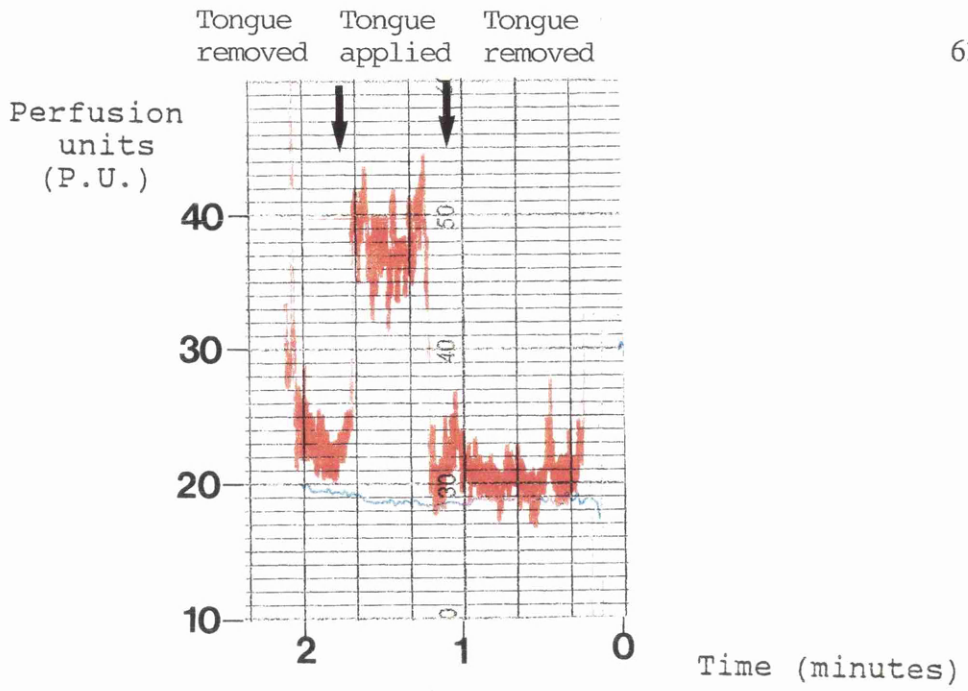


Figure 3.8 The effect on the flux signal recorded from a vital maxillary incisor when the patient placed their tongue against the palatal surface of the crown during the recording.

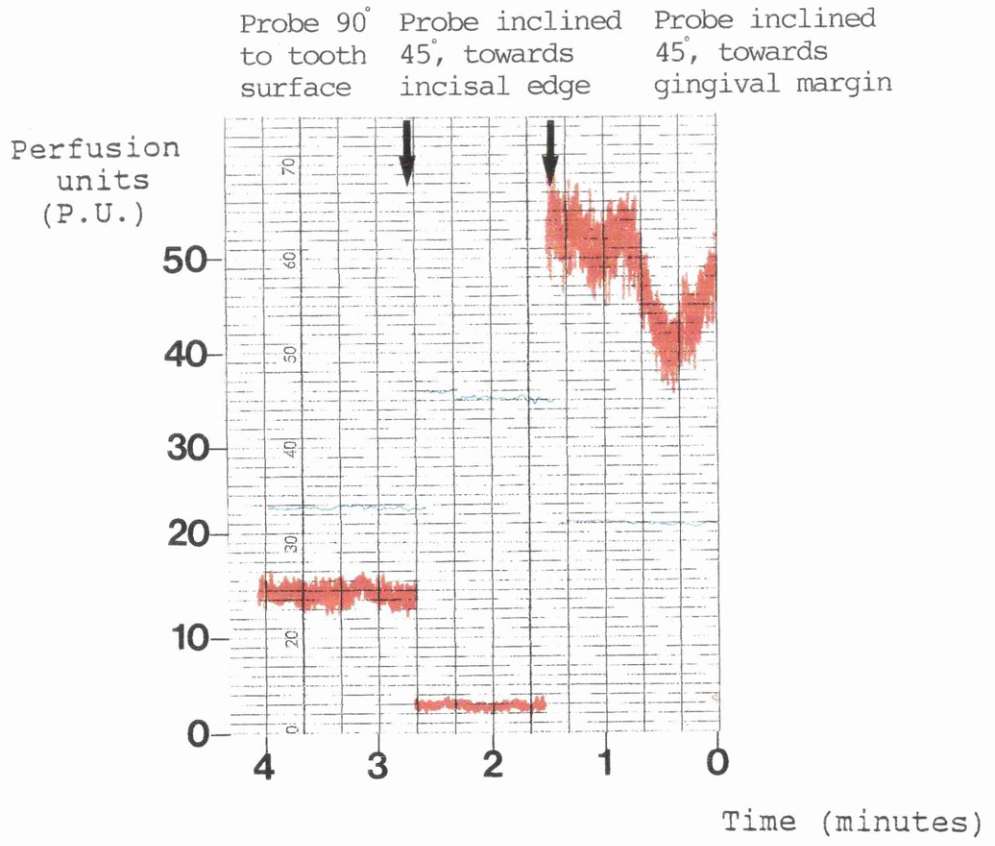


Figure 3.9 The effect on the flux signal recorded from a vital maxillary incisor of altering the angle between the fibre-optic probe and the tooth surface.

Table 3.1 Percentage measurement error in repeat visual assessments of 50 chart recordings of pulpal flux signals.

Flux signal variable	Mean % change (SD)	Range
Mean Flux	1.9% (2.6%)	0-6.9%
Amplitude of Slow Wave Vasomotion	17.8% (15.9%)	0-85.7%
Amplitude of Cardiac Cycle	14.9% (14.3%)	0-40.0%

Table 3.2. Classification error in assessment of frequency bands of Slow Wave Vasomotion

Charts reclassified	No change in frequency band	Changed ± 1 frequency band	Changed ± 2 frequency bands	Changed ± 3 frequency bands
N=50	44%	42%	12%	2%

from a maxillary incisor recorded over a period of 30 minutes shown in Figure 3.10. As the amplitude of Slow Wave Vasomotion can alter the Mean Flux value by up to 100% (Figure 3.10) a minimum recording period must include at least one full cycle and should, therefore, extend for at least one minute. Wilder-Smith (1988), in an investigation of the effect of restorative materials on pulpal blood flow using laser doppler flowmetry, only recorded the flux signal for 10 seconds.

In the present study, Slow Wave Vasomotion was quantified by measuring the amplitude of the three most representative consecutive cycles during a recording. Recordings off the incisal edge of non-vital teeth (Figure 3.4) show Slow Wave Vasomotion of amplitude 1.0 P.U., due to instrument and recording noise, and this value is, therefore, used as the minimum value for Slow Wave Vasomotion in all analyses. A reported value of 1.0 P.U., therefore, indicates that physiological Slow Wave Vasomotion was not identified. To assess measurement error, 50 recordings were remeasured after one month and the results are presented in Table 3.1. The periodicity of Slow Wave Vasomotion was quantified from the time period for the three cycles identified for amplitude assessment, and from that obtaining the average time period for one cycle. The times noted were assigned to the nearest of the following intervals; 13, 20, 27, 33, 40, 47 and 53 seconds per cycle. To assess measurement error, 50 recordings were remeasured after an interval of one month. The results are shown in Table 3.2.

3.5.4 Cardiac Cycle signal

The Cardiac Cycle signal is the flux signal frequency which corresponds with the cardiac cycle. It can be identified between A-B in Figure 3.3. This signal was found to be synchronous with the cardiac cycle as recorded by a finger mounted pulse oximeter. Two variables can be identified in the Cardiac Cycle signal; its amplitude and its regularity. In the present study, Cardiac Cycle signal amplitude was assessed visually from the chart recording. Measurement error was determined by reassessing

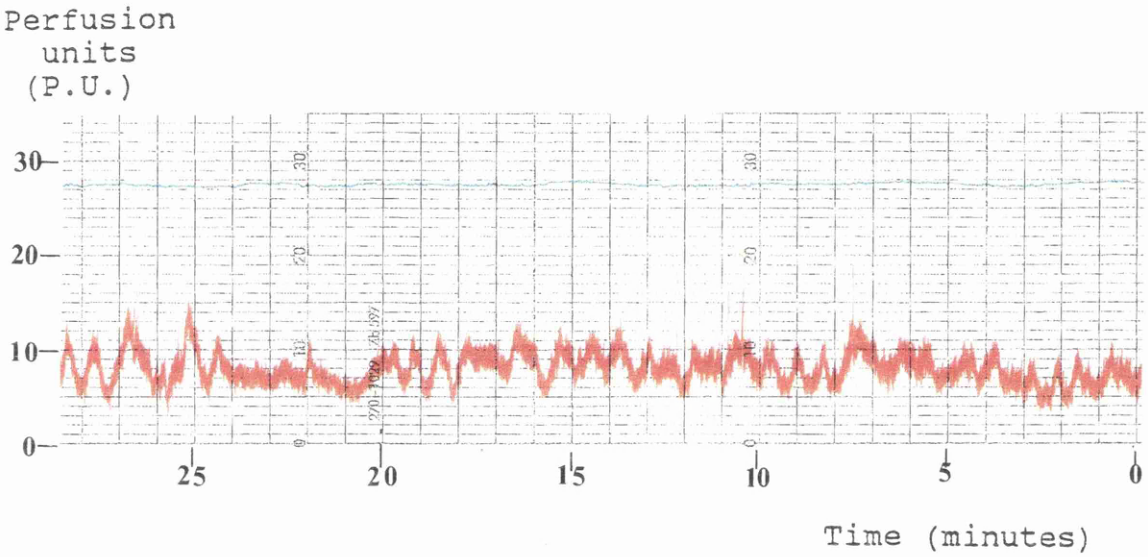


Figure 3.10 Flux signal recorded from a vital maxillary incisor over a period of 30 minutes.

50 charts after one month, and the results are shown in Table 3.1. Although the Cardiac Cycle signal originates from fluctuations in pulpal perfusion due to the pulsatile nature of blood flow, the signal does not always show a regular cardiac pulse, even when originating from a vital dental pulp (see Section 5.2). Nevertheless, regularity of the cardiac cycle signal may be useful in pulpal diagnosis (Olgart *et al.*, 1988) and this will be investigated in Chapter 6. If the Cardiac Cycle signal was regular, it was classified as having a cardiac pulse present. Alternatively a cardiac pulse was classified as being equivocal or absent. To assess classification error in assessing the presence of a cardiac pulse signal, 239 chart recordings from both vital and non-vital dental pulps were reclassified randomly and blind after an interval of at least one month. The results are shown in Table 3.3.

3.5.5 Discussion

The assessment of the flux signal will form the basis of any study using laser doppler flowmetry and it is essential that the repeatability of the assessment method is quantified. The results of repeating the measurement of Mean Flux and amplitude of Slow Wave Vasomotion after one month are shown in Table 3.1, and are shown as percentage error. This was calculated by dividing the difference between two readings by their mean, expressed as a percentage. The lowest measurement error was for Mean Flux, with a mean percentage error of only 1.9% and a maximum error of 7% in reassessing 50 recordings. Although Slow Wave Vasomotion was identified in all 50 recordings on both measuring occasions, quantification of this flux signal variable was not as reliable as for Mean Flux, with a mean percentage error of 17.8% (SD 15.9%). Classifying the frequency of Slow Wave Vasomotion was particularly unreliable. Less than half the 50 charts were reclassified into the same frequency band (see Table 3.2) and this variable was, therefore, not included in further analyses. The relatively poor reliability of visual methods of measuring Slow Wave Vasomotion was probably due to the irregularity that can occur in the frequency of this flux signal

Table 3.3. Classification error in repeat assessments of presence of Cardiac Pulse signal in 239 chart recordings.

Original classification	Reclassified as present	Reclassified as equivocal	Reclassified as absent
Present (n=153)	97%	3%	0%
Equivocal (n=31)	22%	52%	26%
Absent (n=55)	0%	15%	85%

variable, causing difficulty in identifying the three most representative cycles within a recording. However, it is possibly the presence or absence of Slow Wave Vasomotion rather than its amplitude, which is of importance in pulpal diagnosis using laser doppler flowmetry (Olgart *et al.*, 1988).

Assessing the amplitude of the Cardiac Cycle signal was slightly less prone to error than assessing the amplitude of Slow Wave Vasomotion, with a mean percentage error of 14.9% (SD 14.4%) but was still not as reliable as assessing Mean Flux. As might be anticipated, classification of the Cardiac Cycle regularity showed the lowest error when a cardiac pulse was originally classified as either present (only 3% reclassified differently) or absent (15% reclassified differently). Half of the recordings with equivocal regularity were reclassified differently. However, equivocal recordings only made up 13% of the recordings in this sample. As there was a greater tendency for an absent cardiac pulse to be reclassified as equivocal (15%) than there was for a regular cardiac pulse to be reclassified as equivocal (3%), equivocal cardiac pulse signals were grouped with absent cardiac pulse signals for the rest of the study.

In summary, visual assessment of Mean Flux would appear to be a reliable method of quantifying this flux signal variable. Visual assessment of the amplitude of Slow Wave Vasomotion may be subject to measurement error and this should be born in mind when defining absolute values as diagnostic criteria for this variable. An initial assessment of a Cardiac Cycle signal as having a cardiac pulse present is likely to be reliable. In view of the inherent measurement error in visual analysis of the chart recordings, the possibility of using computer aided analysis was explored and this will now be discussed.

3.6 A STUDY ON FOURIER TRANSFORMATION OF THE FLUX SIGNAL

3.6.1 Introduction

Frequency distribution within a signal output may be investigated using Fourier transformation. This mathematical process is well known although to date there have

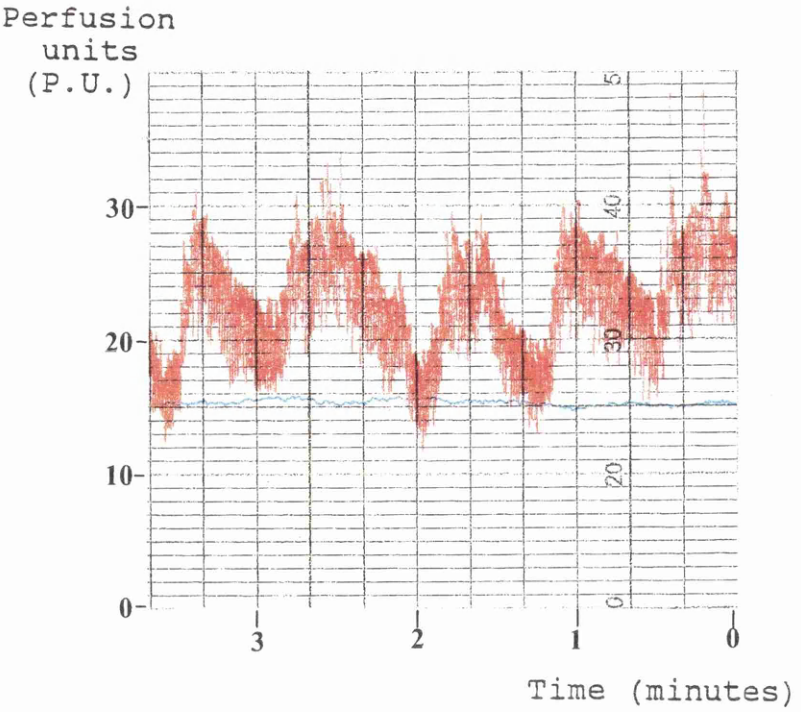
been no reports in the literature of its use in analysing the flux signal from the dental pulp. Fourier transformation quantifies a signal in terms of its frequency components. A computer program was developed by the Department of Computing Science at the University of Strathclyde which allowed frequency analysis of the pulpal flux signal using Fourier transformation. The flux signal output from the flowmeter was digitised using an 'A to D' board and the data transferred to an IBM PC. The computer analysis was based on a three minute recording, with a sampling rate of ten flux voltage readings per second. The mean voltage of the recording over the three minutes was classified as flux signal variable V, and was the direct equivalent of Mean Flux. The amplitude of frequencies between 1-10 cycles per minute (equivalent to Slow Wave Vasomotion frequencies) were presented as flux signal variable V2. The amplitude of frequencies between 11-55 cycles per minute were presented as flux signal variable V3 and those over 55 cycles a minute (equivalent to the cardiac cycle frequencies) as flux signal variable V4. The total amplitude of all the frequencies during the recording were calculated as flux signal variable V1. A chart recording of a typical pulpal flux signal, and the graphical representation of the frequency distribution within the same signal obtained using Fourier transformation, are shown in Figure 3.11.

The aim of this study was to investigate the correlation between the flux frequency variables V, V1, V2, V3 and V4 and the flux signal variables Mean Flux, amplitude of Slow Wave Vasomotion and amplitude of the Cardiac Cycle signal.

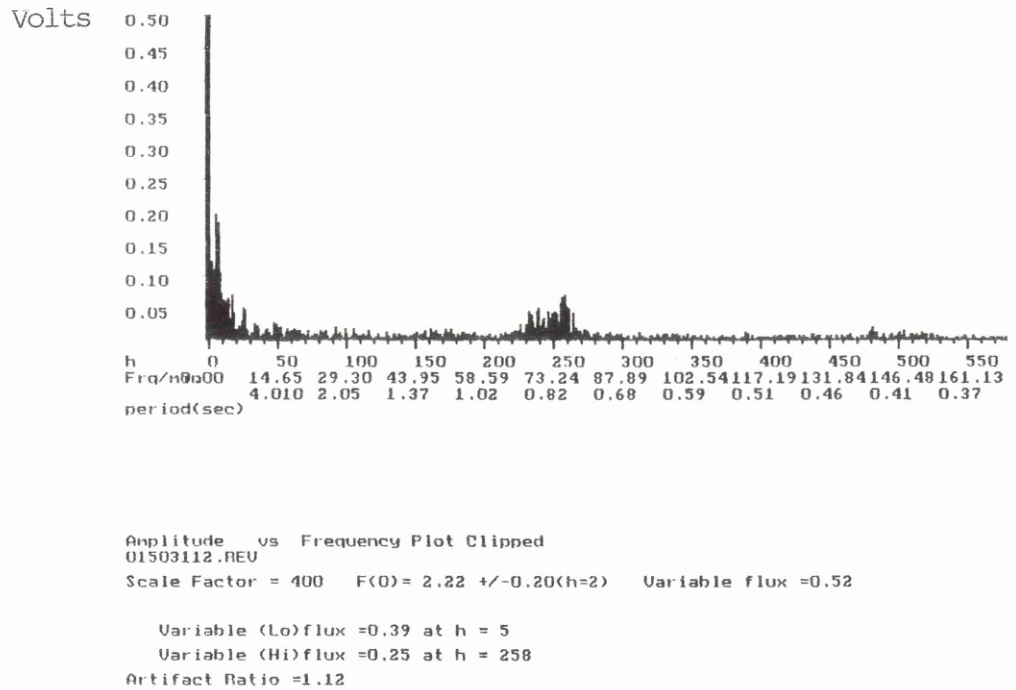
3.6.2 Materials and method

The sample consisted of 140 flux signal recordings from vital permanent maxillary canines and incisors, and mandibular incisors from 10 young adult volunteers (5 females, 5 males, mean age 24 years 4 months (range 22-29 years)). The recording method was as described in Appendix A. The data were analysed visually and using Fourier transformation. Flux signal variables assessed were Mean

a)



b)



Source of flux signal frequency variables V and V1-V4 (see Section 3.6):-

F(0)= V +/- V2
Variable flux= V1
Variable (Lo) flux= V3
Variable (Hi) flux= V4

Figure 3.11 The flux signal from a vital maxillary incisor (a), and at (b) a computer analysis of the frequency distribution within the same signal produced using Fourier transformation.

Flux, amplitude of Slow Wave Vasomotion, amplitude of the Cardiac Cycle, mean flux signal voltage V, and the four frequency groupings of V1, V2, V3 and V4 which were defined in Section 3.6.1. Correlation between the flux signal variables was analysed using a statistical software package (Minitab, 7.2) on an I.B.M. personal computer. The significance of the correlation was assessed.

3.6.3 Results

The results for this study are shown in Table 3.4. The highest level of correlation was between Mean Flux and flux frequency variable V (0.99) and the lowest was between the amplitude of Slow Wave Vasomotion and amplitude of the Cardiac Cycle (0.28). However, all the flux signal variables were significantly correlated with each other to some degree. Correlation coefficients in the range 0.25-0.29 were significant at the level of $p<0.05$, and coefficients >0.29 were significant at the level of $p<0.001$.

3.6.4 Discussion

The results show that all flux signal variables were significantly correlated with each other at least at the level $p<0.05$. However, it should be noted that, in view of the large numbers in the sample, a significant correlation does not necessarily imply good agreement. For example, Figure 3.12 shows a dotplot of Flux frequency variable V1 against V2; good agreement reflected by the correlation coefficient of 0.99. Figure 3.13 shows a dotplot of flux frequency variable V2 against V4; comparatively much reduced agreement but still with a correlation coefficient of 0.38, and still significant at the level of $p<0.001$. The highest level of correlation was found between Mean Flux and mean flux voltage V at 0.99. This would be expected as both are measures of the same parameter. However, the result does support the validity of measuring Mean Flux visually as does the low error in repeating measurements of this parameter reported in Table 3.1. Cardiac Cycle amplitude and flux frequency group

Table 3.4. Correlation coefficients between flux signal variables from 140 pulpal flux recordings.

	Amp SWV	Amp. CC	V	V1	V2	V3	V4
Mean Flux	0.43	0.66	0.99	0.59	0.37	0.63	0.82
Amp. SWV		0.28	0.41	0.73	0.61	0.77	0.40
Amp. CC			0.65	0.46	0.31	0.35	0.82
V				0.58	0.35	0.63	0.82
V1					0.93	0.84	0.59
V2						0.63	0.38
V3							0.57

Amp. SWV- Amplitude of Slow Wave Vasomotion
Amp. CC - Amplitude of Cardiac Cycle

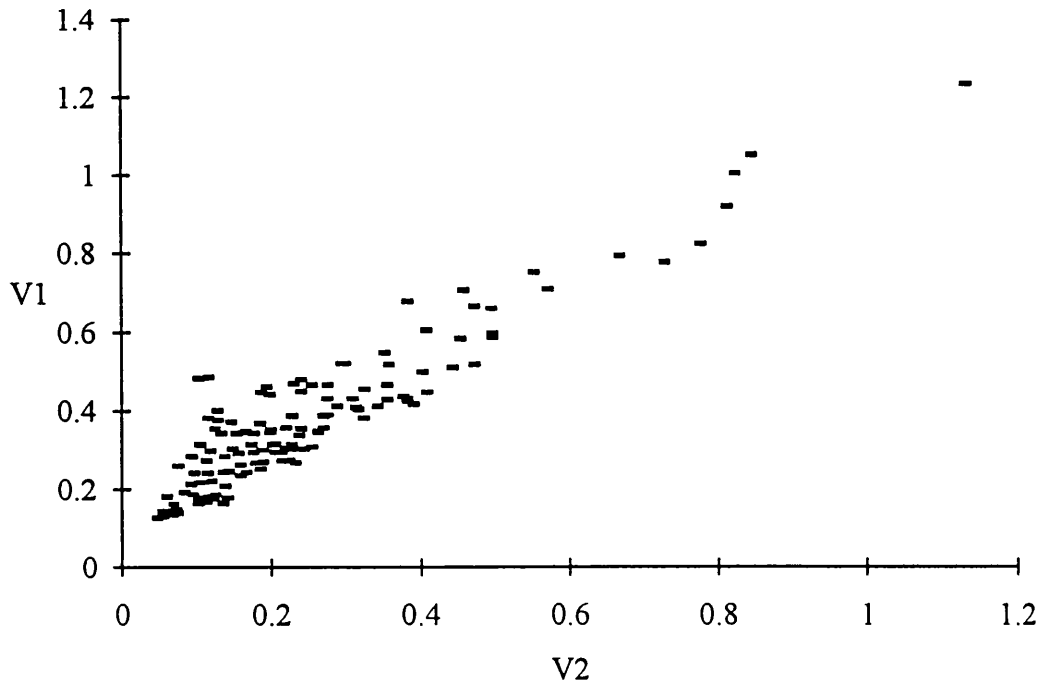


Figure 3.12 Plot of flux frequency variable V1 against V2 for 140 pulpal flux recordings.
Correlation Coefficient = 0·93 Significance P < 0·001

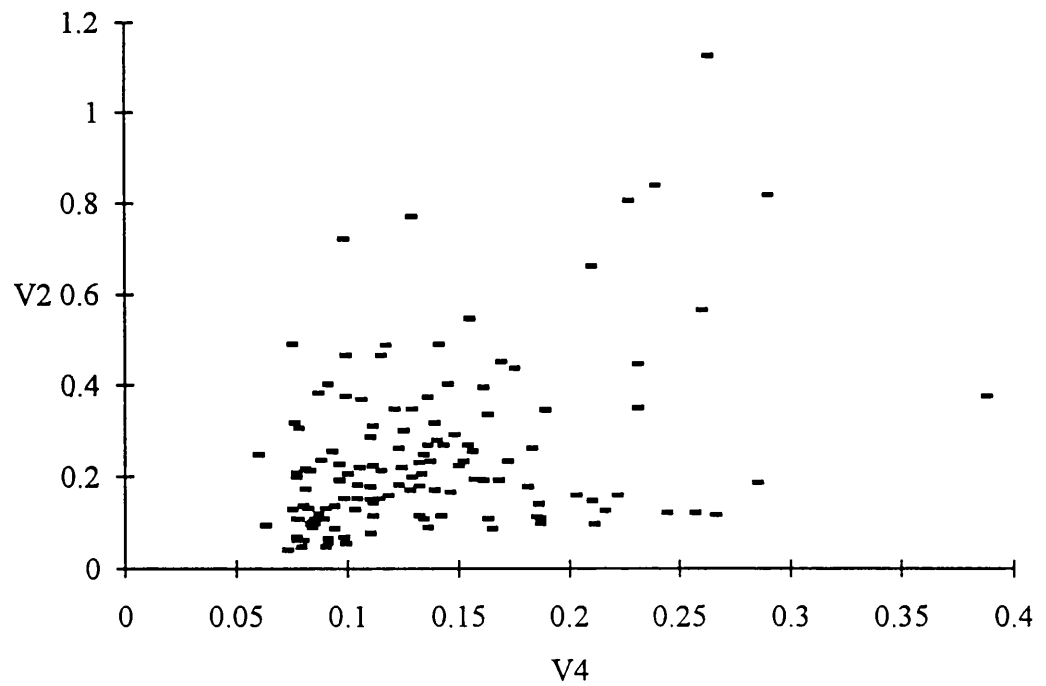


Figure 3.13 Plot of flux frequency variable V2 against V4 for 140 pulpal flux recordings.
Correlation Coefficient = 0·38 Significance P < 0·001

V4 also had a high correlation coefficient (0.82), again due to both being measures of the same underlying parameter. Mean Flux was highly correlated (0.82) with flux frequency grouping V4, which could be anticipated due to the cardiac cycle being the single most important source of kinetic energy within the circulatory system; the stronger the pulse, the greater the blood flow. What was a little more surprising was that amplitude of Slow Wave Vasomotion was more highly correlated with the medium frequency grouping V3, at 0.77, than the low frequency grouping V2, at 0.61. This may reflect difficulties in accurately measuring the amplitude of Slow Wave Vasomotion visually, a method which is prone to error (Section 3.5.3).

3.6.5 Summary and conclusion

The aim of this thesis is to develop laser doppler flowmetry as a method of assessing the vitality of the traumatised dental pulp. Perfect correlation between the flux signal variables described would indicate that only one, and indeed any one, variable would be of use in reaching a diagnosis. However, although all the variables were significantly related to each other the correlation between them was not perfect and, therefore, several flux signal variables in conjunction may prove to be of use in reaching a diagnosis of pulpal status. These will be discussed in Chapter 6. Having discussed the recording parameters for laser doppler flowmeters and the analysis of the flux signal output, the development of a reliable recording method for laser doppler flowmetry of the dental pulp will be described in Chapter 4.

CHAPTER 4

DEVELOPMENT OF METHOD FOR RECORDING PULPAL BLOOD FLOW USING LASER DOPPLER FLOWMETRY

4.1 INTRODUCTION AND AIMS

Pulpal blood flow is recorded using laser doppler flowmetry by placing a fibre-optic probe against the tooth surface. Several methods of holding the probe have been described in the literature and these are summarised in Table 4.1. However, a potential problem in attempting to measure pulpal blood flow using laser doppler flowmetry is the recording of non-pulpal signals. These signals could arise from non-pulpal blood flow, such as from the peridontium, and also from non blood flow signals caused by probe movement.

The dental pulp is enclosed within a calcified crystalline shell, which itself is surrounded by highly perfused peridontium and it is, therefore, likely that an optical method of recording pulpal blood flow will include a proportion of non-pulpal blood flow within the measurement. This is acceptable when using laser doppler flowmetry to discriminate between vital and non-vital dental pulps, provided that the proportion of the signal due to non-pulpal blood flow is low and remains constant. However, non-pulpal blood flow becomes more problematic when laser doppler flowmetry is used to investigate changes in pulpal blood flow, as it is known that the physiological control of pulpal blood flow and gingival blood flow are different (Aars *et al.* 1992b; Watson *et al.* 1992). In addition, laser doppler flowmeters will detect doppler induced frequency shifts in light if the probe is moving relative to the reflector. It is, therefore, essential that a valid recording technique both screens gingival tissue from the laser doppler flowmeters recording volume and adequately stabilises the fibre-optic probe against the tooth surface.

The optimum recording method for pulpal blood flow is one which minimises the proportion of non-pulpal signals within the flux signal. One way of assessing this is the non-pulpal/pulpal flux signal ratio. However, only the Swedish group (Gazelius *et al.* 1986 & 1988; Olgart *et al.* 1988) and Wilder-Smith (1988) have published data allowing computation of the non-pulpal/pulpal flux signal ratio (Table 4.1) for the recording method used. The proportion of a reported pulpal flux signal which is actually of non-pulpal origin cannot be determined without this data.

The aim of this chapter was to develop the recording method to be used in this thesis. Due to the paucity of published data validating the various recording methods it was necessary to investigate the non-pulpal flux signal for the various methods described. The study was carried out in two stages; firstly, the optimum method of probe fixation was investigated and secondly, the optimum spatial positioning of the probe using that method of probe fixation was determined.

4.2 INVESTIGATION OF METHODS OF LASER DOPPLER FLOWMETER PROBE FIXATION.

4.2.1 Introduction and aims.

A variety of methods for holding the laser doppler flowmeter probe against the tooth surface have been reported in the literature and these are summarised in Table 4.1.

This study had the following aims:-

a) to determine the level of non-pulpal flux signal for the various recording methods reported in the literature

b) to develop a recording technique which minimised the non-pulpal flux signal

c) to determine the non-pulpal/pulpal flux signal ratios for comparison with other studies

Table 4.1. A review of the recording techniques used in published studies on laser doppler flowmetry of the human dental pulp.

Ratio NV:V - Ratio of Mean Flux signal from non-vital/vital dental pulps (N=number in sample)

Publication	Laser source	Bandwidth filter	Probe stabilisation	Ratio NV:V
Gazelius *et al.* 1986	633 nm	Narrow	Rubber dam and clamp	0.17 (n=1)
Olgart *et al.* 1988	633 nm	Narrow	Elastomeric impression	0.13 (n=33)
Gazelius *et al.* 1988	633 nm	Narrow	Elastomeric impression	0.07 (n=4)
Wilder-Smith 1988	633 nm	Narrow	Hand held	0.22 (n=5)
Vongsavan *et al.* 1991	810 nm	Wide	Hard acrylic splint	Not stated
Ramsay *et al.* 1991a	633 nm	Wide	Labial impression	Not stated
Ramsay *et al.* 1991b	633 nm	Wide	Soft vinyl splint	Not stated
Petterson *et al.* 1991	750 nm	Not stated	Hand Held	Not stated
Watson *et al.* 1992	780 nm	Not stated	Polythene splint	Not stated
Aars *et al.* 1992	633 nm	Narrow	Elastomeric impression	Not stated
Ingolfsson *et al.* 1993	633 nm	Narrow	Elastomeric impression	Not stated
Pitt Ford *et al.* 1993	800 nm	Not stated	Elastomeric impression	Not stated

4.2.2 Materials and methods

The study compared recordings of pulpal flux signals from vital and non-vital dental pulps with seven different methods of laser doppler flowmeter probe fixation. The study sample comprised 17 non-vital and 16 vital maxillary incisor teeth from a population of 17 patients (mean age 13 years 5 months, range 7 years 7 months to 20 years 4 months) attending Glasgow Dental Hospital following dental trauma. A dental pulp was classified as vital if the patient responded to pulpal sensibility testing and there were no clinical signs of pulpal necrosis. A dental pulp was classified as non-vital if there was no response to pulpal sensibility testing and there were at least two other clinical signs of pulpal necrosis (radiographic signs, coronal discolouration, tenderness to percussion). Laser doppler flowmetry of the dental pulp is time consuming and as the study was carried out on patients attending for treatment, there was insufficient time within an appointment to use all seven recording methods on each study tooth. A pilot study indicated that the full impression technique (see Appendix A for a complete description) gave the lowest non-pulpal flux signal. This method was, therefore, used as a control, each tooth having a pulpal flux recording made with this method and one or more alternative methods. The mean number of incisors in each vital teeth and non-vital teeth recording groups was six (range 5-9). The laser doppler flowmetry recordings were taken using the PF2b's narrow 4 KHz waveband filter, with the gain at the maximum setting of 100 times. The minimum recording period was one minute. The laser doppler flowmeter's probe was positioned at the junction of the gingival third and the middle third of the labial surface of the crown of the maxillary incisor. The recording method used on every tooth in the study was the full impression technique. This was a modification of the method described by Gazelius *et al.* (1988). A disposable plastic impression tray was trimmed to include only the six anterior teeth. A putty and wash elastomeric impression (Provil, Bayer) of the maxillary teeth was then taken and holes prepared to hold the laser doppler flowmeter's fibre-optic probe perpendicular to the labial surface of the crown of the study tooth. In addition, further recordings were taken with one or more of the following methods;

(a) Hand held; the probe was held by hand (Wilder-Smith, 1988; Pettersson & Oberg, 1991). (5 vital, 9 non-vital teeth).

(b) Hand held + rubber dam; the probe was held by hand and the tooth isolated with green rubber dam to provide screening from gingival blood flow. (5 vital, 8 non-vital teeth).

(c) Tube jig; the probe was inserted into a 1 cm length of 2 mm diameter orthodontic tubing which was retained perpendicular to the tooth surface with bonding resin. (7 vital, 5 non-vital teeth).

(d) Tube jig + rubber dam; the probe was positioned as in method (c) with green rubber dam providing screening from gingival blood flow (Gazelius *et al.* 1986). (5 vital, 5 non-vital teeth).

(e) Soft vinyl splint; a clear thermoplastic soft vinyl splint was fabricated on a study model, and a hole through which the probe was inserted was prepared perpendicular to the labial surface of the tooth (Ramsay *et al.* 1991b). (9 vital, 6 non-vital teeth).

(f) Labial impression; a putty and wash elastomeric impression (Provil, Bayer) was taken of the anterior maxillary teeth using a stock tray. The elastomer was then removed from the tray and trimmed so that only the labial face remained, with some overlap of the incisal edge of the teeth to allow location. Holes were prepared to hold the probe perpendicular to the labial surface of the tooth (Ramsay *et al.* 1991a). (6 vital, 7 non-vital teeth).

The number of recordings taken from maxillary incisors categorised as non-vital and from those categorised as vital are indicated in brackets following the description of the method. The non-pulpal/pulpal flux signal ratio was calculated by dividing the mean value of Mean Flux obtained from the non-vital pulps by the mean value of Mean Flux obtained from the vital teeth.

4.2.3 Results

The Mean Flux signals recorded from the subject teeth using the recording methods described in Section 4.2.2 (a-f) and the full impression method as an intra-tooth control, are shown in Tables 4.2 (a-f).

Table 4. 2 (a) Mean Flux values from non-vital and vital teeth using the hand held and full impression recording methods

a) Non-vital teeth

Subject tooth	Mean Flux (P.U) Hand held	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	20	3	-85.0
2	15	1	-93.3
3	47	2	-95.7
4	19	1	-94.7
5	18	6	-66.7
6	14	6	-57.1
7	30	4	-86.7
8	19	3	-84.2
9	15	2	-86.7
Mean (SD)	21.9 (10.5)	3.1 (1.9)	-83.4 (13.1)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Hand Held	Mean Flux (P.U.) Full Impression	Percentage change using Full Impression
1	80	54	-32.5
2	35	18	-48.6
3	42	24	-42.9
4	50	35	-30.0
5	33	25	-24.2
Mean .(SD)	48.0 (19.1)	31.2 (14.1)	-35.6 (9.9)

Ratio of means of Mean Flux non-vital/vital teeth	
Hand held	0.46
Full Impression	0.10

Table 4.2 (b) Mean Flux values from non-vital and vital teeth using the hand held + rubber dam method and the full impression recording method

a) Non-vital teeth

Subject tooth	Mean Flux (P.U.) Hand Held + Rubber Dam	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	7	3	-57.1
2	10	1	-90.0
3	19	2	-89.5
4	7	1	-85.7
5	13	6	-53.8
6	20	4	-80.0
7	19	3	-84.2
8	7	2	-71.4
Mean (SD)	12.8 (5.8)	2.8 (1.7)	-76.5 (14.3)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Hand held + Rubber dam	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	41	54	-31.7
2	39	24	+38.5
3	34	30	+11.8
4	30	35	-16.7
5	15	25	-66.7
Mean (SD)	31.8 (10.3)	33.6 (12.2)	-13.0 (40.3)

Ratio of means of Mean Flux non-vital/vital teeth	
Hand held + Rubber dam	0.40
Full impression	0.08

Table 4.2 (c) Mean flux values from non-vital and vital teeth using the tube jig and full impression recording methods

a) Non-vital teeth

Subject tooth	Mean Flux (P.U.) Tube jig	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	13	3	-76.9
2	10	1	-90.0
3	12	4	-66.7
4	8	3	-62.5
5	20	5	-75.0
Mean (SD)	12.6 (4.6)	3.2 (1.5)	-74.2 (10.6)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Tube jig	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	54	54	0
2	44	31	-29.5
3	28	17	-39.3
4	25	23	-8.0
5	62	37	-40.3
6	38	35	-7.9
7	54	38	-29.6
Mean (SD)	43.6 (14.0)	33.6 (11.9)	-22.1 (16.5)

Ratio of means of Mean Flux non-vital/vital teeth	
Tube jig	0.29
Full impression	0.09

Table 4.2 (d) Mean Flux values from non-vital and vital teeth using the tube jig and rubber dam , and the full impression recording methods

a) Non-vital teeth

Subject tooth	Mean Flux (P.U.) Tube jig + rubber dam	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	6	3	-50.0
2	1	1	0
3	6	4	-33.3
4	4	3	-25.0
5	10	5	-50.0
Mean (SD)	5.4 (3.3)	3.2 (1.5)	-31.7 (20.8)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Tube jig + rubber dam	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	50	54	+8.0
2	25	17	-32.0
3	16	23	+43.8
4	36	35	-2.8
5	49	38	-22.4
Mean (SD)	35.2 (14.9)	33.4 (14.4)	-1.1 (29.6)

Ratio of means of Mean Flux non-vital/vital teeth	
Tube jig + rubber dam	0.15
Full impression	0.10

Table 4.2 (e) Mean Flux values from non-vital and vital teeth using the vinyl splint and full impression recording methods

a) Non-vital teeth

Subject tooth	Mean Flux (P.U.) Vinyl splint	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	7	3	-57.1
2	15	2	-86.7
3	11	1	-90.9
4	11	2	-81.8
5	8	2	-75.0
6	15	4	-73.3
Mean (SD)	11.2 (3.4)	2.3 (1.0)	-77.5 (12.0)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Vinyl splint	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	50	54	+8.0
2	37	21	-43.2
3	27	18	-33.3
4	22	15	-31.8
5	27	23	-14.8
6	22	10	-54.5
7	11	10	-9.1
8	21	12	-42.9
9	50	38	-24.0
Mean (SD)	29.7 (13.4)	22.3 (14.7)	-27.3 (19.5)

Ratio of means of Mean Flux non-vital/vital teeth	
Vinyl splint	0.38
Full impression	0.10

Table 4.2 (f) Mean Flux values from non-vital and vital teeth using the labial impression and full impression recording methods

a) Non-vital teeth

Subject tooth	Mean Flux (P.U.) Labial impression	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	5	3	-40.0
2	16	4	-75.0
3	15	2	-86.7
4	6	1	-83.3
5	11	6	-45.5
6	6	4	-33.3
7	10	3	-70.0
Mean (SD)	9.9 (4.5)	3.3 (1.6)	-62.0 (21.9)

b) Vital teeth

Subject tooth	Mean Flux (P.U.) Labial impression	Mean Flux (P.U.) Full impression	Percentage change using full impression
1	35	19	-45.7
2	29	14	-51.7
3	32	24	-25.0
4	42	30	-28.6
5	42	26	-38.1
6	54	38	-29.6
Mean (SD)	39.0 (9.0)	25.2 (8.4)	-36.5 (10.6)

Ratio of means of Mean Flux non-vital/vital teeth	
Labial impression	0.25
Full impression	0.13

4.2.4 Discussion

The mean value of Mean Flux fron non-vital teeth when the probe was held by hand (21.9 P.U., SD 10.5) fell to 12.6 P.U. (SD 4.5) when the tube jig was used, with a further fall to 5.4 P.U.(SD 3.2) when the tube jig was used in conjunction with rubber dam. This suggests a compound effect of probe movement and non-pulpal blood flow as potential components of a pulpal blood flow signal. These components were minimised by using the full impression technique, which consistently gave lower mean values for Mean Flux from non-vital teeth (range 3.1-2.7 P.U.) with comparatively small standard deviations (range 1.9-1.0 P.U.) when compared with the alternative techniques.

The full impression technique generally produced lower mean values for Mean Flux from vital teeth than the other recording methods, but this reduction was always less than the reduction in mean values of Mean Flux recorded from non-vital teeth. This resulted in the full impression technique having the lowest non-pulpal/pulpal Mean Flux signal ratio of all the methods tested.

The results show that although all the methods of holding the laser doppler flowmeter probe allowed the detection of pulpal blood flow there was considerable variation in the proportions of non-pulpal flux signal obtained. When the probe was hand held, nearly 50% of the flux signal obtained was of non-pulpal origin. Probe stabilisation alone, such as provided by the soft vinyl splint, tube jig and labial impression probe holding methods, still resulted in 25-38% of a pulpal flux signal being of non-pulpal origin. The source of this non-pulpal blood flow was most likely to have been the peridontium, as screening the gingival margin with rubber dam reduced the flux signal from both vital and non-vital teeth when the probe was hand held and when the tube jig was used. Both Ingolfsson *et al.* (1993) and Aars *et al.* (1992b) noted that gingival blood flow might contribute to the pulpal flux signal, but neither author investigated this further.

In the present study the lowest non-pulpal flux signal was obtained using the full impression technique. With this technique it may be deduced that only about 10% of the signal obtained from a vital dental pulp is of non-pulpal origin. Although the full impression technique required the longest chairside time to use (with the exception of

the soft vinyl splint technique which involved laboratory work) it provided both probe stabilisation and some screening of the periodontal tissues from the measuring volume of the laser doppler flowmeter. When the full impression method was used to record from the incisal edge of a non-vital maxillary incisor the flux signal output was equivalent to the signal from an extra-oral static reflector used to obtain a zero flux (Figure 3.4). It would, therefore, seem that the flux signal obtained from non-vital teeth using the full impression technique originated almost entirely from non-pulpal blood flow, and not from instrument noise or probe/tooth movement. In view of these findings the full impression recording technique was used for all further work in this thesis.

4.2.5 Conclusion

Studies using laser doppler flowmetry to measure pulpal blood flow must minimise the non-pulpal component of the flux signal. When the laser doppler flowmeter probe was placed at the junction of the gingival third and middle third of the labial surface of the dental crown (the position used in previous studies (Section 4.3.2)), signals of non-pulpal origin were an unavoidable component of flux recordings from the dental pulp.

A useful indicator of non-pulpal signals is the non-pulpal/pulpal flux signal ratio for the recording method used. Without this information the proportion of the reported pulpal flux signal which is actually of periodontal origin cannot be determined. For the PF2b laser doppler flowmeter, with a laser source of 633 nm, the lowest non-pulpal/pulpal flux signal ratio (0.10) was obtained using the full impression recording method.

4.3 INVESTIGATION OF THE SPATIAL POSITIONING OF THE LASER DOPPLER FLOWMETER PROBE

4.3.1 Introduction and aims

The study of recording methods indicated that perfused non-pulpal tissue could be included within the measuring volume of the laser doppler flowmeter when the probe was applied to a tooth surface. It would seem likely that the major source of

non-pulpal blood flow was the peridontium, and that the proportion of the signal from this source could be reduced by increasing the separation of the probe from the gingival margin. However, the volume of dental pulp tissue within the crown decreases with increasing distance from the gingival margin, due to the morphology of the pulp chamber, and this would be expected to result in a fall in pulpal flux signal. In addition, the clinical crown length may be reduced due to loss of tooth tissue following crown fracture, incomplete eruption of an immature tooth or following intrusive luxation. It would be clinically advantageous to be able to record pulpal blood flow as near to the gingival margin as possible.

With the exception of Wilder-Smith (1988) and Pettersson and Oberg (1991), neither of whom stated a recording distance, all the studies listed in Table 4.1 used a distance of between 2-3 mm separation of the probe from the gingival margin. However, only Ramsay *et al.* (1991b) investigated the effect of varying the position of the probe on pulpal flux values, and then only on vital maxillary central incisors. The permanent anterior teeth vary in hard tissue morphology and gingival contour and, therefore, the optimum recording distance might vary with tooth type.

The aims of this study were;

a) to determine the optimum separation of the laser doppler flowmeter probe from the gingival margin of anterior teeth for minimising the non-pulpal/pulpal flux signal ratio.

b) to pilot study the criteria for discriminating between flux signals from vital and non-vital dental pulps, which will then be investigated further in a subsequent study.

4.3.2 Materials and methods

Recordings of pulpal blood flow were taken from non-vital anterior teeth with the laser doppler flowmeter probe at 1 mm, 2 mm and 3 mm from the gingival margin. These recordings were compared with recordings from vital anterior teeth made following the same methodology. It would have been preferable to use vital teeth from the same group of patients who provided the non-vital teeth. There were, however, two factors which made this aim difficult to achieve. Firstly, patients who

had sustained dental trauma of sufficient severity to devitalise a tooth may have sustained some injury to the contra-lateral tooth, particularly with regard to maxillary and mandibular central incisors. Secondly, recording at three probe positions on one tooth required the fabrication of a second full impression jig. This was to ensure complete separation of the laser doppler flowmeter probe holes. One jig was used for the 2 mm recording and the other for the 1 mm and 3 mm recordings. This, in conjunction with the additional recordings, added to an already lengthy appointment and difficulties were encountered with patient and parent compliance. The vital tooth sample, therefore, comprised 12 young adult volunteers (mean age 23 years 6 months, range 21-30 years) with no history of dental trauma and with anterior teeth which were either unrestored, or minimally restored, and which were classified as vital according to the criteria described in Section 4.2.2. Using the recording method described in Appendix A, recordings were taken from 12 maxillary and mandibular central incisors, 11 maxillary lateral incisors and 11 maxillary canine teeth. Due to recording difficulties, data were unavailable from 1 maxillary canine and 1 maxillary lateral incisor, each from a different volunteer.

The non-vital tooth sample comprised 20 non-vital, non-endodontically treated, anterior teeth in 18 patients (mean age 14 years, range 9 years 6 months-24 years 6 months) attending Glasgow Dental Hospital following dental trauma. The teeth were classified non-vital using the criteria outlined in Section 4.2.2. Of the 20 teeth classified as non-vital, 12 were subject to pulpectomy within 2 weeks of laser doppler flowmetry and all were found to be clinically non-vital. Recordings of pulpal blood flow were taken from the following groups of non-vital teeth; maxillary central incisors (n=8), maxillary lateral incisors (n=5), maxillary canines (n=2) and mandibular incisors (n=5). The low number of maxillary canines in the sample reflects the low proportion of these teeth affected by trauma.

Recordings were taken from the vital and non-vital teeth with the laser doppler flowmetry probe at 1 mm, 2 mm and 3 mm from the gingival margin on each tooth. The distance of the probe hole from the gingival margin in the impression jig was measured with a William's pattern periodontal probe. Variables contained within the

flux signal were assessed using the methodology described in Section 3.5. For each recording the flux signal variables described were Mean Flux, presence of Slow Wave Vasomotion and presence of a cardiac pulse. Individual Mean Flux values obtained at the three probe positions were compared by two-way analysis of variance. Where significant differences were detected, pairs of positions were compared using paired t tests. As a data set was used for more than one analysis, the level of significance was adjusted to $p<0.017$ following Bonferroni correction (Kleinbaum, Kupper & Muller, 1988). Slow Wave Vasomotion was described as present if its amplitude exceeded 1 P.U., otherwise it was described as absent. A cardiac pulse signal was described as present or absent.

4.3.3 Results

The Mean Flux values obtained at the three probe positions are shown in Table 4.3 and in Figure 4.1. The non-pulpal/pulpal flux signal ratios derived from these values are shown in Table 4.3 and in Figure 4.2. The Mean Flux signals from both vital and non-vital teeth were found to decrease with increased separation from the gingival margin. Statistical analysis was not carried out on the Mean Flux values obtained from non-vital maxillary canines due to the small sample size (n=2). For the remainder of the sample the Mean Flux values obtained at 3 mm probe separation from the gingival margin were significantly less than those obtained at 1 mm probe separation for all tooth types, both vital and non-vital, at least at the level of $p<0.017$. The greatest reduction in the non-pulpal/pulpal flux signal ratio occurred when the laser doppler flowmeter probe separation from the gingival margin increased from 1 mm to 2 mm. There was a smaller reduction when moving from 2 mm to 3 mm except for mandibular incisors when there was a small increase.

The percentage of flux signals from the grouped samples of non-vital teeth (n=18) and vital teeth (n=46) demonstrating Slow Wave Vasomotion at the different

Table 4.3 Variation in Mean Flux and non-vital/vital flux signal ratio from vital and non-vital dental pulps with probe separation from gingival margin.

SD - Standard Deviaton PU - Perfusion Units

Maxillary Central Incisor

Probe Sep.	Flux (PU) (SD) Vital N=12	Non-Vital N=8	Ratio NV/V
1 mm	32.9 (14.8)	9.0 (4.0)	0.27
2 mm	22.9 (7.4)	3.2 (1.9)	0.14
3 mm	17.5 (8.5)	2.2 (1.3)	0.13

Maxillary Lateral Incisor

Probe Sep.	Flux (PU) (SD) Vital N=11	Non-Vital N=5	Ratio NV/V
1 mm	38.9 (13.8)	16.8 (7.1)	0.36
2 mm	30.3 (12.6)	3.0 (1.0)	0.10
3 mm	22.3 (12.4)	1.6 (1.3)	0.07

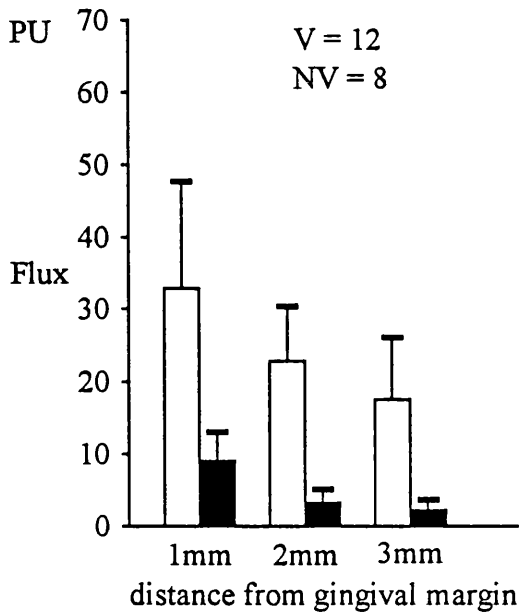
Maxillary Canine

Probe Sep.	Flux (PU) (SD) Vital N=11	Non-Vital N=2	Ratio NV/V
1 mm	29.1 (7.4)	7.5 (2.1)	0.26
2 mm	17.1 (6.3)	2.5 (0.7)	0.15
3 mm	10.6 (4.8)	1.5 (2.1)	0.14

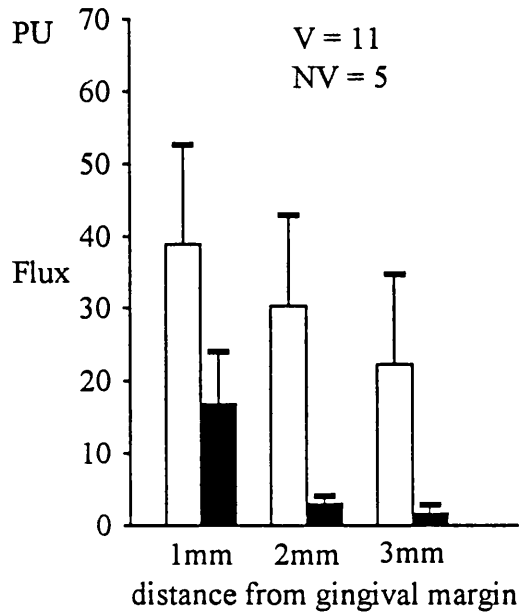
Mandibular Central Incisor

Probe Sep.	Flux (PU) (SD) Vital N=12	Non-Vital N=5	Ratio NV/V
1 mm	43.7 (19.2)	18.4 (7.8)	0.42
2 mm	29.6 (14.8)	3.4 (1.1)	0.12
3 mm	20.0 (14.3)	3.0 (1.2)	0.15

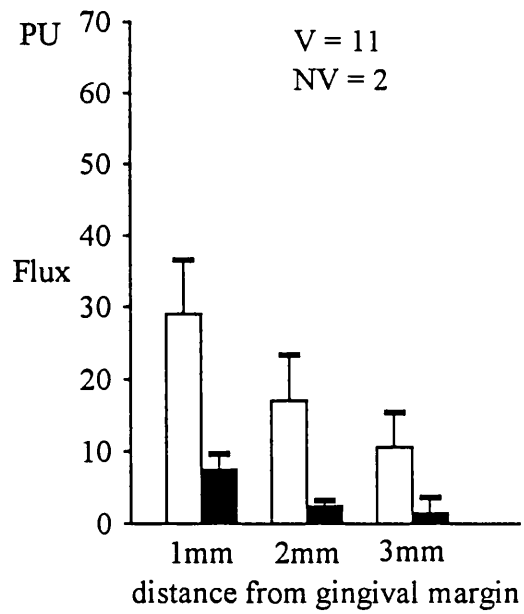
Maxillary Central Incisor



Maxillary Lateral Incisor



Maxillary Canine



Mandibular Incisor

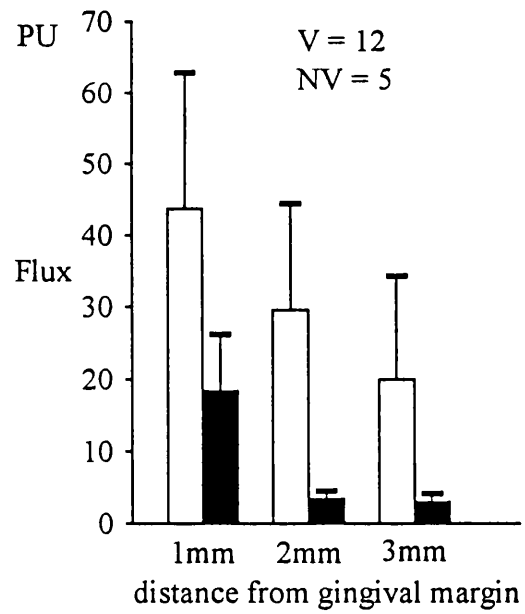


Figure 4.1 Variation of Mean Flux signal from vital and non-vital dental pulps with variation in probe separation from the gingival margin

□ Vital ■ Non-vital ⊤ one standard deviation PU - Perfusion Units

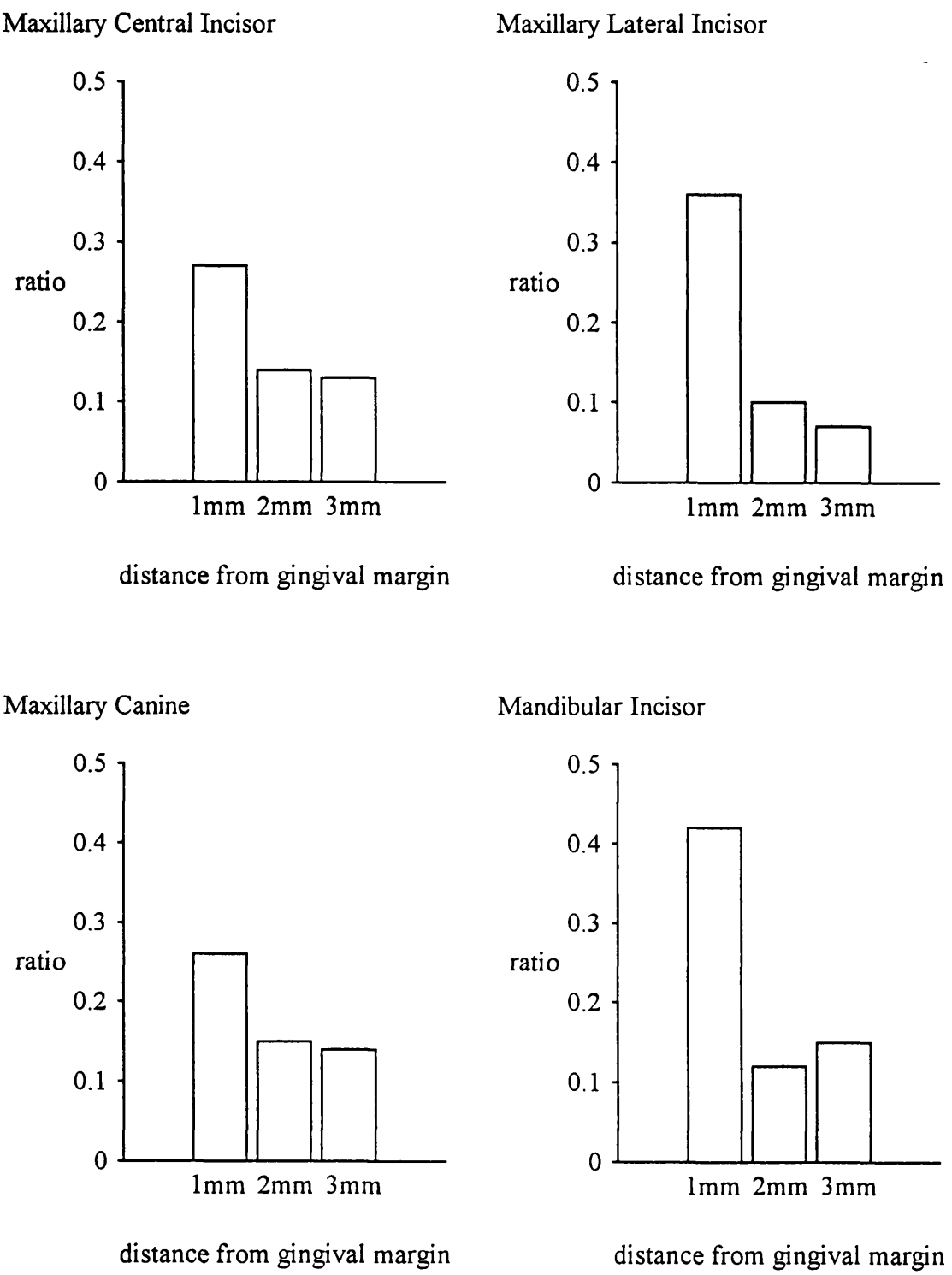


Figure 4.2 Ratio of Mean Flux signal from non-vital pulps against vital pulps with variation in probe separation from the gingival margin

probe positions are shown in Table 4.4. The percentage of flux signals from the grouped samples of non-vital teeth (n=19) and vital teeth (n=46) demonstrating a regular cardiac pulse signal at different probe positions are shown in Table 4.5.

4.3.4 Discussion

The finding that the flux signal from vital dental pulps was significantly related to probe position in the long axis of the tooth, increasing in value as the gingival margin was approached, is in agreement with the findings of Ramsay *et al.* (1991b). The increase in flux signal at 1 mm from the gingival margin may be the result of the increased volume of dental pulp tissue beneath the probe, due to the morphology of the dental pulp chamber. However, the flux signal from non-vital dental pulps was also significantly increased at 1 mm from the gingival margin, indicating that periodontal blood flow contributed to the signal from the dental pulp at this distance.

The ratios of non-vital/vital pulpal flux signals in Table 4.3 show that the minimum ratio was obtained at 2 mm from the gingival margin for mandibular incisors and at 3 mm from the gingival margin for maxillary anterior teeth. However, the reduction in the ratio from 2 mm to 3 mm for the maxillary teeth was small in comparison with the reduction gained from moving the recording distance from 1 mm to 2 mm from the gingival margin.

In deciding the optimum probe position other factors have to be considered. The possibility that the absence of the flux signal variable of Slow Wave Vasomotion might indicate loss of dental pulp vitality was discussed in Section 3.5.3. Table 4.4 shows that Slow Wave Vasomotion was absent from all recordings from non-vital dental pulps (n=18) at a recording distances of 3 mm from the gingival margin, but was present in 5% of recordings at 2 mm and 72% of recordings taken at 1 mm from the gingival margin. Although Slow Wave Vasomotion was present in 98% of recordings from vital dental pulps at 1 mm and 92% of recordings at 2 mm from the gingival margin, it was only present in 60% of recordings taken at 3 mm. The optimum probe position for using Slow Wave Vasomotion as a diagnostic criterion would, therefore, seem to be 2 mm from the gingival margin.

Table 4.4 Percentage of flux signals from vital and non-vital anterior teeth showing Slow Wave Vasomotion (SWVM), with probe separation from gingival margin.

	Percentage of teeth with SWVM	
Distance from gingival margin	Vital N = 46	Non-vital N = 18
1mm	98	72
2mm	92	5
3mm	60	0

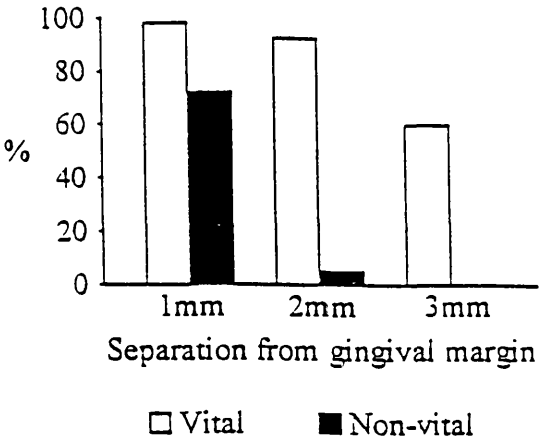
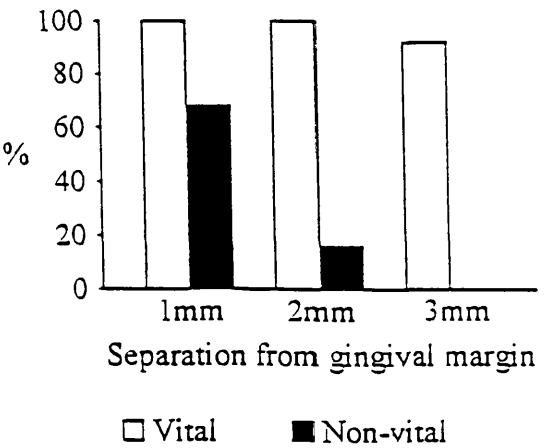


Table 4.5 Percentage of flux signals from vital and non-vital anterior teeth having a regular cardiac pulse (RCP), with probe separation from the gingival margin.

	Percentage of teeth with RCP	
Distance from gingival margin	Vital N = 46	Non-vital N = 19
1mm	100	68
2mm	100	16
3mm	92	0



The possibility that absence of a regular cardiac pulse within the Cardiac Cycle signal might indicate loss of dental pulp vitality was discussed in Section 3.5.4. Table 4.5 shows that the cardiac pulse signal was present in 68% of recordings (n=19) from non-vital dental pulps taken 1 mm from the gingival margin. Increasing the probe separation to 2 mm reduced the percentage to 16%, with all recordings from vital dental pulps having a regular cardiac pulse at this distance. Although at 3 mm no flux signal from non-vital teeth had a regular cardiac pulse, it was also absent from 8% of flux signals from vital pulps at this distance. The optimum probe position for using cardiac pulse regularity would seem to be 3 mm from the gingival margin.

Assessment of all the factors discussed indicated that for a study involving the classification of pulp vitality using laser doppler flowmetry, the optimum probe/gingival margin separation was 2 mm. As the impression jig is prepared by hand at the chairside some degree of error in positioning the probe hole is inevitable. This study indicated that if error in preparing the probe hole was to occur, then it should be on the side of increasing probe separation from the gingival margin above 2 mm. Because of this the optimum recording distance is defined as being between 2 mm and 3 mm from the gingival margin.

To assess error in positioning the probe hole, study models were subsequently prepared from 43 impression jigs used to record pulpal blood flow from 77 teeth and the probe hole/gingival margin separation measured with Vernier gauge orthodontic callipers. The sample was found to have a mean probe/gingival margin separation of 2.3 mm (SD 0.24, range 1.8-3.0 mm).

A review of the range of values for the flux signal variables Mean Flux and Slow Wave Vasomotion obtained from non-vital teeth at 2-3 mm from the gingival margin indicated that the majority of the Mean Flux values were <7 Perfusion Units (P.U.), and that Slow Wave Vasomotion was usually absent but that if present was unlikely to have an amplitude >1.5 P.U.. These values formed the basis of the laser doppler flowmetry diagnostic criteria investigated in Chapter 6.

4.3.5 Conclusion

The optimum recording technique for investigating pulpal blood flow using laser doppler flowmetry was to use a two-stage elastomeric impression to hold the fibre-optic probe perpendicular to the tooth surface between 2-3 mm from the gingival margin.

CHAPTER 5

LASER DOPPLER FLOWMETRY OF THE VITAL DENTAL PULP

5.1 INTRODUCTION AND AIMS OF CHAPTER

The hypothesis tested in this thesis is that, in the vitality assessment of the dental pulp of traumatised permanent incisors, laser doppler flowmetry (L.D.F.) has a sensitivity and specificity better than other diagnostic methods in current use. The diagnosis of disease depends on recognising deviation from normal and the study reported in Chapter 4 indicated that normal flux signals recorded by laser doppler flowmetry from vital dental pulps had two characteristics: Mean Flux with a minimum value of 7 perfusion units (P.U.) and Slow Wave Vasomotion with a minimum amplitude of 1·6 P.U.. It is, therefore, possible that these values (hereafter referred to as the L.D.F. classification criteria) might be used as diagnostic criteria to discriminate between flux signals from vital and non-vital dental pulps. However, Mean Flux values recorded from vital dental pulps in the study showed wide variation: the coefficient of variation (standard deviation/mean) in the various recording groups ranging between 0.33-0.65. Variation in the values of flux signal variables recorded from different teeth and the repeatability of flux signal variables recorded from the same tooth on different occasions are both factors which might affect the validity of laser doppler flowmetry used to investigate changes in dental pulp blood flow. Large variations in flux signal variables between patients would cause problems with inter-patient comparisons while poor repeatability of flux signal variables recorded from the same tooth on different occasions would cause problems with intra-patient comparisons. However, in the present study laser doppler flowmetry was used only to discriminate between dental pulps where blood flow was either present or absent. Variation in the values of flux signal variables recorded from dental pulps and poor repeatability of flux signal variables would, therefore, only become problematic if

sufficient to result in those values crossing the L.D.F. classification criteria threshold, resulting in the flux signal being incorrectly classified.

The aim of this chapter was to investigate the normal range of values for flux signal variables from vital anterior teeth and to determine if the L.D.F classification criteria were likely to result in a vital dental pulp being incorrectly classified as non-vital. The first part of the study investigated the inter-patient variability of flux signal variables obtained under standardised recording conditions and also compared the flux signal variables recorded from different anterior tooth types. The second part of the study investigated the intra-patient repeatability of flux signal variables obtained both under standardised recording conditions and with variations in recording conditions which might be unavoidable in a longitudinal clinical study.

5.2 AN INVESTIGATION OF THE NORMAL RANGE OF FLUX SIGNAL VARIABLES FROM VITAL ANTERIOR TEETH.

5.2.1 Introduction and aims.

The aim was to investigate the normal range of flux signal variables for vital anterior teeth, to determine whether these values differed between tooth types and between patients, and to determine if the range of values fell below the L.D.F. classification criteria described in Section 5.1.

5.2.2 Materials and Method.

The study group comprised ten adults (5 females, 5 males) mean age 24 years 4 months (range 22-29 years). Ideally, the sample should have comprised patients of mean age 11 years 6 months (range 6.5 - 24.5 years) to match with the sample of patients included in the study on traumatised anterior teeth reported in Chapter 7. However, for reasons of practicability; to exclude patients with traumatised teeth and the need for subjects to attend for appointments of over 1.5 hours, it was decided to use young adult volunteers. The study group comprised 10 junior house staff. The teeth selected were intact or with minimal restorations, had no history of traumatic injury and responded positively to sensibility testing. The laser doppler recording

method used is described in Appendix A. Recordings were taken at the same time of day (late afternoon), with the subjects rested, supine and starved for the previous two hours. Recordings were taken from the maxillary canines (13)(23), lateral incisors (12)(22) and central incisors (11)(21) and from the mandibular central incisors (41)(31), in random order and in a single recording session, from each of the 10 volunteers. The flux signal output was quantified as described in Section 3.5. Flux signal variables quantified were Mean Flux, amplitude of Slow Wave Vasomotion, amplitude of Cardiac Cycle and flux signal frequency variables V1, V2, V3 and V4. In addition, the presence of a cardiac pulse within the cardiac cycle signal was noted.

Percentage differences between two flux signal variables (from pairs of teeth from the same volunteer) were calculated as the difference between the variables divided by their mean value. The coefficient of variation for a sample was calculated by dividing the standard deviation by the mean. Differences in flux signal variables between the four groups of teeth were investigated using analysis of variance. Where differences were identified, these were investigated using least significant difference multiple range tests.

5.2.3 Results

A total of 77 recordings of pulpal blood flow, recorded from eight anterior teeth in each of ten patients, were analysed; three recordings of lower incisors (from three different patients) were not used due to recording difficulties caused by incisor imbrication. For each patient, recordings were taken from all the teeth during a single session. The means and standard deviations of flux signal variables for the individual tooth types investigated are shown in Table 5.1 (a-d) and in Figure 5.1 (a-g). The presence of a regular cardiac pulse within the Cardiac Cycle signal is shown in Table 5.2.

Table 5.1 (a). Flux signal variables from vital maxillary central incisors

Amp. SWV = Amplitude Slow Wave Vasomotion CC = Cardiac Cycle
P.U. = Perfusion Units V = Volts (teeth n = 20, patients n = 10)

Flux signal variable	Mean Flux (SD)	Range	Coefficient of variation	Percentage difference in range
Mean Flux (P.U.)	18.25 (5.67)	9.0-31.0	0.31	110
Mean Amp. SWV. (P.U.)	4.10 (2.54)	1.8-12.8	0.62	151
Mean Amp. C.C. (P.U.)	3.65 (1.18)	2.0-6.0	0.32	100
Frequency variable V1.(V)	0.394 (0.152)	0.234-0.822	0.39	111
Frequency variable V2.(V)	0.259 (0.159)	0.096-0.771	0.61	155
Frequency variable V3.(V)	0.219 (0.105)	0.111-0.513	0.48	129
Frequency variable V4.(V)	0.128 (0.038)	0.075-0.209	0.30	94

Table 5.1 (b). Flux signal variables from vital maxillary lateral incisors

Amp. SWV = Amplitude Slow Wave Vasomotion CC = Cardiac Cycle
P.U. = Perfusion Units V = Volts (teeth n = 20, patients n = 10)

Flux signal variables	Mean (SD)	Range	Coefficient of variation	Percentage difference in range
Mean Flux (P.U.)	22.30 (9.66)	7.0-46.0	0.43	147
Mean amp. SWV.(P.U.)	4.35 (1.75)	2.3-8.5	0.40	115
Mean amp. CC.(P.U.)	4.35 (1.93)	2.0-10.0	0.44	133
Frequency variable V1.(V)	0.500 (0.234)	0.178-1.231	0.47	149
Frequency variable V2.(V)	0.343 (0.230)	0.098-1.126	0.67	168
Frequency variable V3.(V)	0.277 (0.106)	0.102-0.453	0.38	126
Frequency variable V4.(V)	0.162 (0.073)	0.082-0.386	0.45	130

Table 5.1 (c) Flux signal variables from vital maxillary canines

Amp. SWV = Amplitude Slow Wave Vasomotion CC = Cardiac Cycle
P.U. = Perfusion Units V = Volts (teeth n=20, patients n=10)

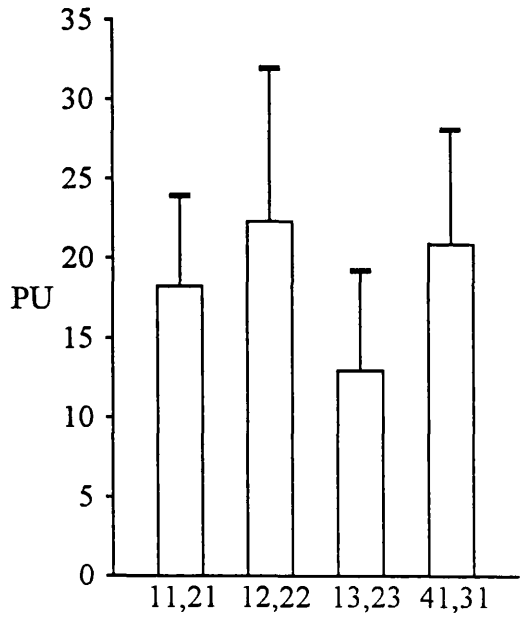
Flux signal Variable	Mean (SD)	Range	Coefficient of variation	Percentage difference in range
Mean Flux (P.U.)	12.95 (6.24)	3.0-30.0	0.48	164
Mean amp. SWV (P.U.)	2.10 (0.84)	1.0-4.1	0.40	122
Mean amp. CC.(P.U.)	2.65 (0.81)	2.0-4.0	0.31	67
Frequency variable V1.(V)	0.207 (0.070)	0.126-0.405	0.34	105
Frequency variable V2.(V)	0.114 (0.063)	0.041-0.310	0.55	153
Frequency variable V3.(V)	0.115 (0.049)	0.054-0.226	0.43	123
Frequency variable V4.(V)	0.088 (0.015)	0.071-0.134	0.17	61

Table 5.1 (d) Flux signal variables from vital mandibular central incisors

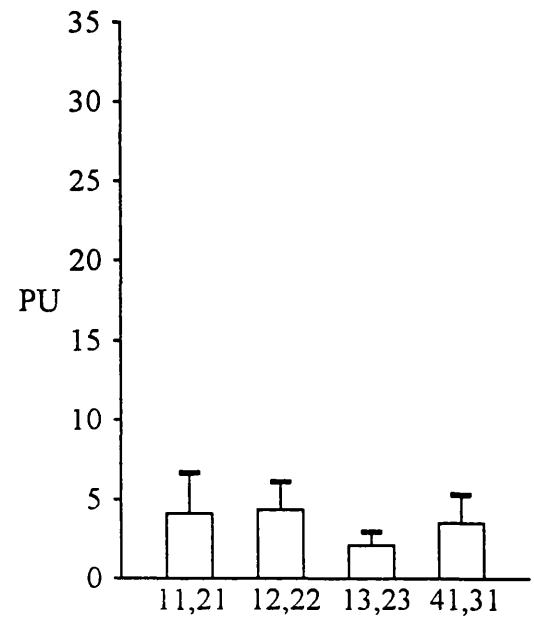
Amp. SWV = Amplitude Slow Wave Vasomotion CC = Cardiac Cycle
P.U. = Perfusion Units V = Volts (teeth n=17, patients n=10)

Flux signal Variable	Mean (SD)	Range	Coefficient of variation	Percentage difference in range
Mean Flux (P.U.)	20.9 (7.2)	10.0-35.0	0.34	111
Mean amp. SWV.(P.U.)	3.52 (1.73)	1.3-9.0	0.49	150
Mean amp. CC.(P.U.)	4.18 (1.18)	2.0-7.0	0.28	111
Frequency variable V1.(V)	0.434 (0.204)	0.191-1.050	0.47	138
Frequency variable V2.(V)	0.263 (0.200)	0.077-0.839	0.76	166
Frequency variable V3.(V)	0.252 (0.120)	0.120-0.576	0.48	131
Frequency variable V4.(V)	0.165 (0.047)	0.098-0.242	0.28	85

a) Mean Flux



b) Mean amplitude of Slow Wave Vasomotion



c) Mean amplitude of Cardiac Cycle

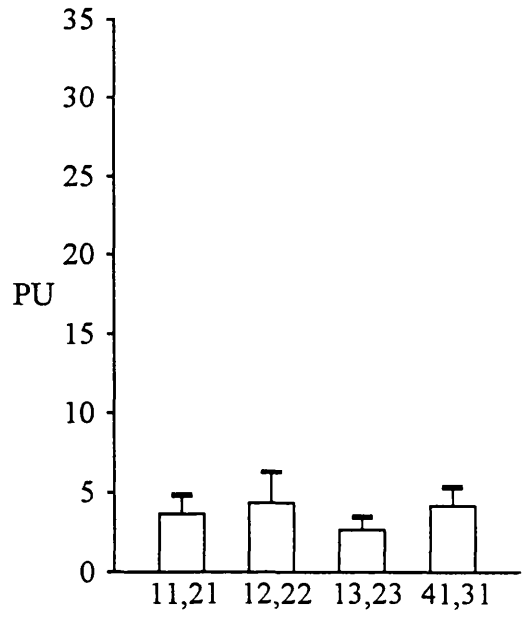
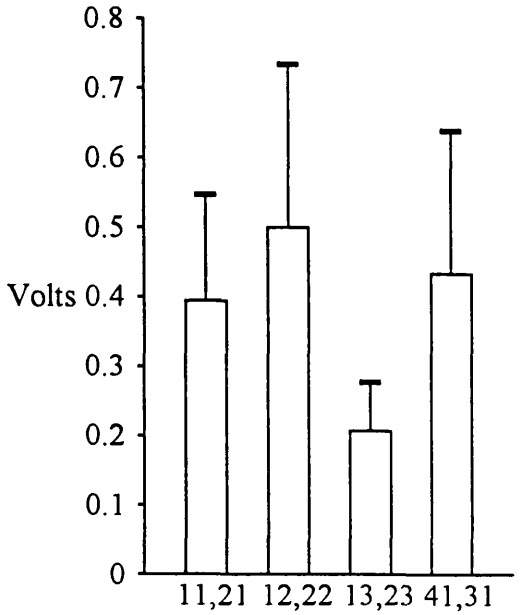


Figure 5.1 (a-g) Flux signal variables by tooth type:-

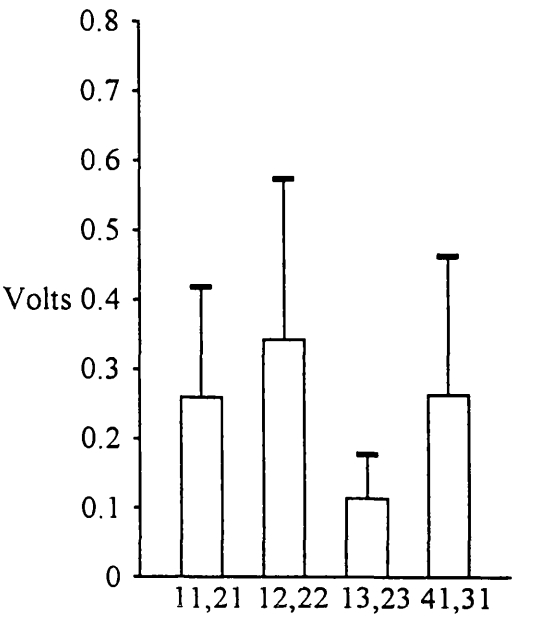
Maxillary Central Incisors (11,21) N = 20 Maxillary Lateral Incisors (12,22) N = 20
Maxillary Canines (13,23) N = 20 Mandibular Central Incisors (41,31) N = 17
Patient Number = 10

⊤ - one standard deviation PU - Perfusion Units

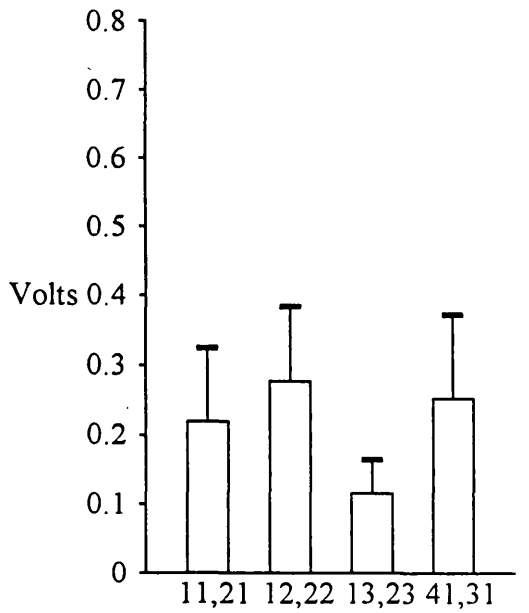
d) Frequency variable V1



e) Frequency variable V2



f) Frequency variable V3



g) Frequency variable V4

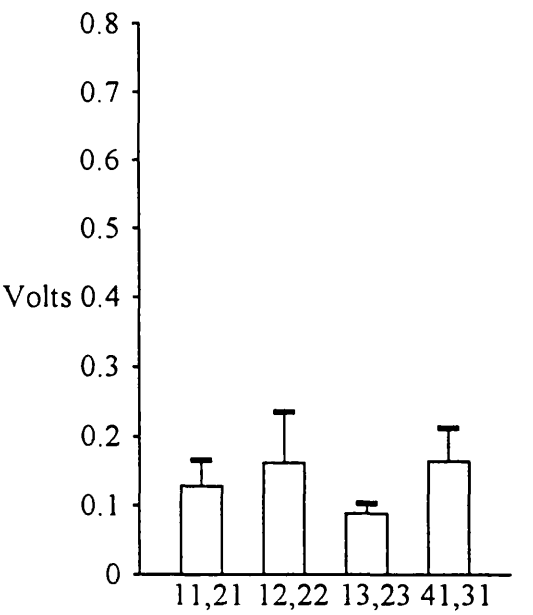


Figure 5.1 Continued; Flux signal variables by tooth type:-

Maxillary Central Incisors (11,21) N = 20 Maxillary Lateral Incisor (12,22) N = 20
Maxillary Canines (13,23) N = 20 Mandibular Central Incisors (41,31) N = 17
Patient Number = 10

T - one standard deviation V - volts

Table 5.2. Percentage of Cardiac Cycle signals containing a regular cardiac pulse.

Tooth type	Maxillary central incisor	Maxillary lateral incisor	Maxillary canine	Mandibular central incisor
Percentage with cardiac pulse	90% (n=20)	95% (n=20)	70% (n=20)	88% (n=17)

The highest mean values for all flux signal variables were obtained from maxillary lateral incisors and the lowest mean values for all variables were obtained from maxillary canines. One way analysis of variance on each flux signal variable indicated a significant difference, at least at the level of $p<0.05$, between the four tooth types for all flux signal variables except for amplitude of the Cardiac Cycle, where no significant difference was found. Further analysis using least sign difference multiple range tests was carried out, and the results are shown in Table 5.3 and in Figure 5.2. For Mean Flux, values obtained from maxillary lateral incisors were significantly higher than from maxillary canines ($p<0.05$), while for amplitude of Slow Wave Vasomotion, values from maxillary canines were significantly lower than from any of the other anterior teeth ($p<0.05$).

The inter-patient variation in flux signal variables from the same tooth types was large, as can be seen from Table 5.1 (a-d). This Table shows the coefficient of variation and range of values for flux signal variables, and in all cases the maximum and minimum values came from different patients. The percentage difference between the maximum and minimum values is also shown. The smallest percentage difference in the range of values of flux signal variables was for the amplitude of the Cardiac Cycle and its associated frequency variable V4, and the lowest of these was for maxillary canines at 61%. Otherwise, the percentage differences in the ranges of values for flux signal variables between the same tooth types in different patients ranged between 110-168%.

The percentage difference in flux signal variables between pairs of teeth of the same tooth type within the same patient are shown in Table 5.4. Although the mean percentage difference in Mean Flux was 25% (SD 19.4%) the range was from 0-88% for this sample of 37 pairs of teeth. The smallest percentage difference in flux signal variables between pairs of teeth was for the amplitude of the Cardiac Cycle signal at 19.6% (SD 22.0%), and the largest was for frequency variable V2 at 52.2% (SD 37.2%). There was no significant difference in any flux signal variable between teeth on the left side of the jaw and their antimeres.

Table 5.3. Least significant difference multiple range analysis of flux signal variables by tooth type.

* = significant difference, at least at $p<0.05$ NS=no significant difference

Maxillary central incisors N=20 Maxillary lateral incisors N=20
Maxillary canines N=20 Mandibular central incisors N=17

Amp. SWV =amplitude of Slow Wave Vasomotion FV=frequency variable
Amp. Card.=amplitude of Cardiac Cycle

a) Maxillary central incisors

	Mean Flux	Amp. SWV	Amp. Card.	FV V1	FV V2	FV V3	FV V4
Maxillary lateral incisors	NS	NS	NS	*	NS	*	*
Maxillary canines	NS	*	NS	*	*	*	*
Mandibular incisors	NS	NS	NS	NS	NS	NS	*

b) Maxillary lateral incisors

	Mean Flux	Amp. SWV	Amp. Card.	FV V1	FV V2	FV V3	FV V4
Maxillary central incisors	NS	NS	NS	*	NS	*	*
Maxillary canines	*	*	NS	*	*	*	*
Mandibular incisors	*	NS	NS	NS	NS	NS	NS

Table 5.3 (cont.). Least significant difference multiple range analysis of flux signal variables by tooth type.

* = significant difference, at least at $p<0.05$ NS=no significant difference

Maxillary central incisors N=20 Maxillary lateral incisors N=20
Maxillary canines N=20 Mandibular central incisors N=17

Amp. SWV =amplitude of Slow Wave Vasomotion FV=frequency variable
Amp. Card.=amplitude of Cardiac Cycle

c) Maxillary canines

	Mean Flux	Amp. SWV	Amp. Card.	FV V1	FV V2	FV V3	FV V4
Maxillary central incisors	NS	*	NS	*	*	*	*
Maxillary lateral incisors	*	*	NS	*	*	*	*
Mandibular incisors	NS	*	NS	*	*	*	*

d) Mandibular incisors

	Mean Flux	Amp. SWV	Amp. Card.	FV V1	FV V2	FV V3	FV V4
Maxillary central incisors	NS	NS	NS	NS	NS	NS	*
Maxillary lateral incisors	*	NS	NS	NS	NS	NS	NS
Maxillary canines	NS	*	NS	*	*	*	*

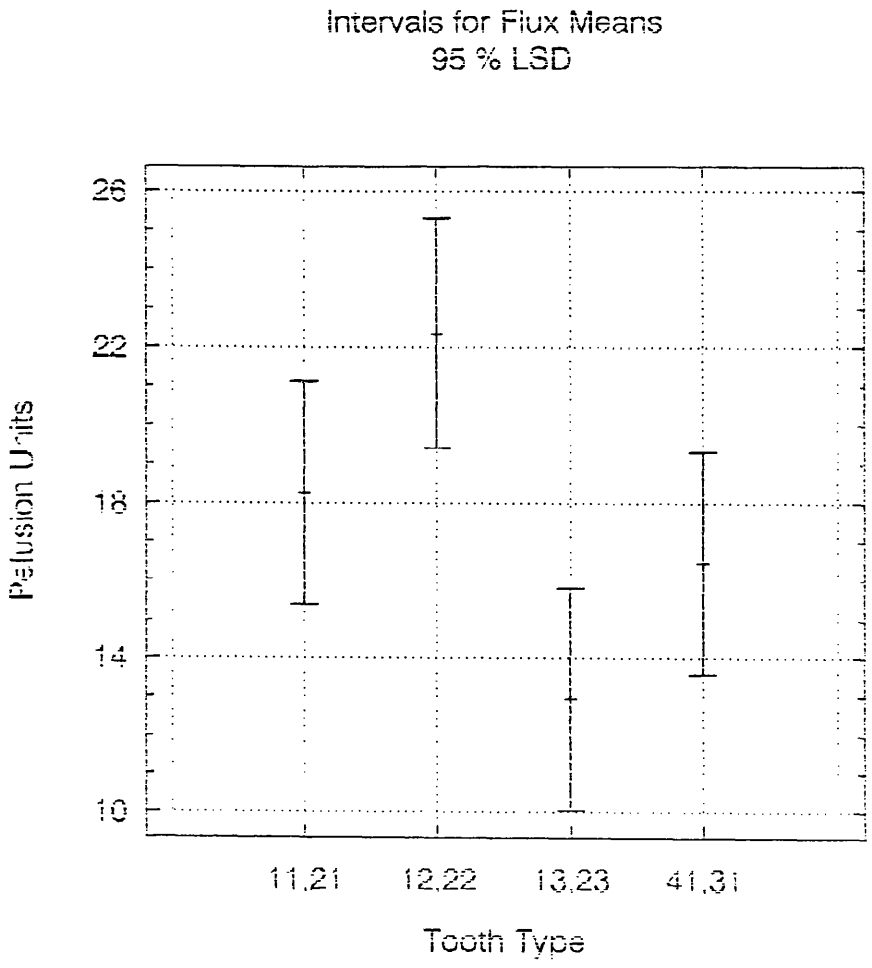


Figure 5.2 (a) 95% least significant difference intervals for Mean Flux by tooth type

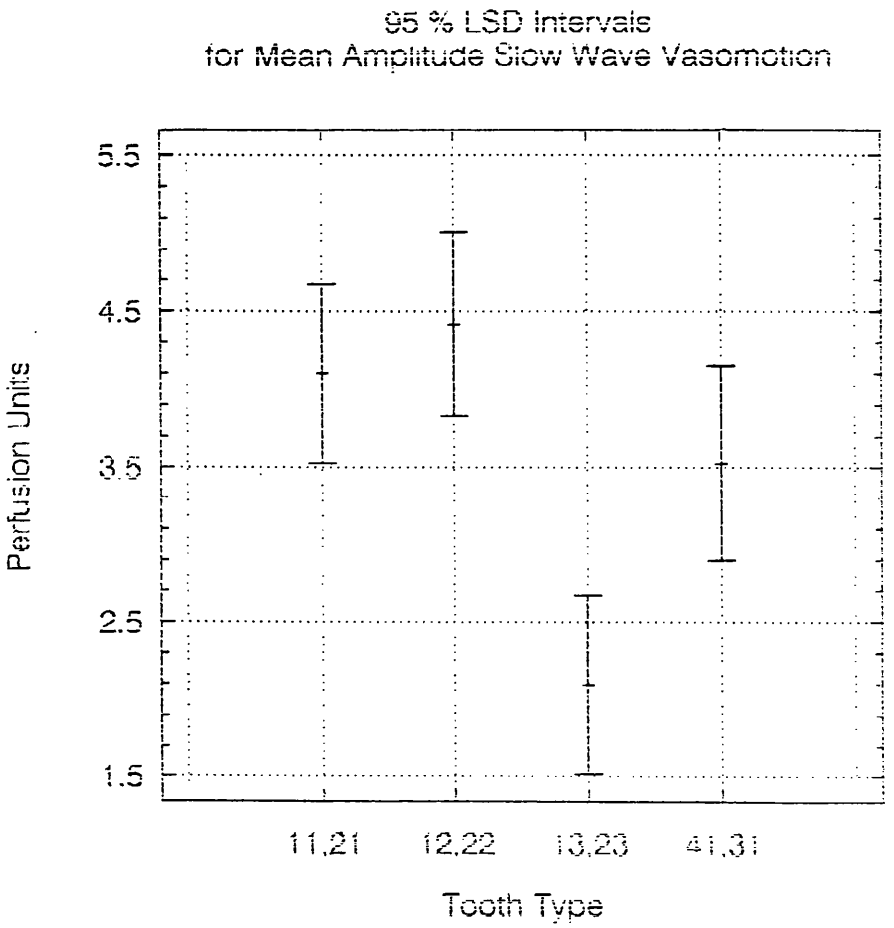


Figure 5.2 (b) 95% least significant difference intervals for amplitude of Slow Wave Vasomotion by tooth type

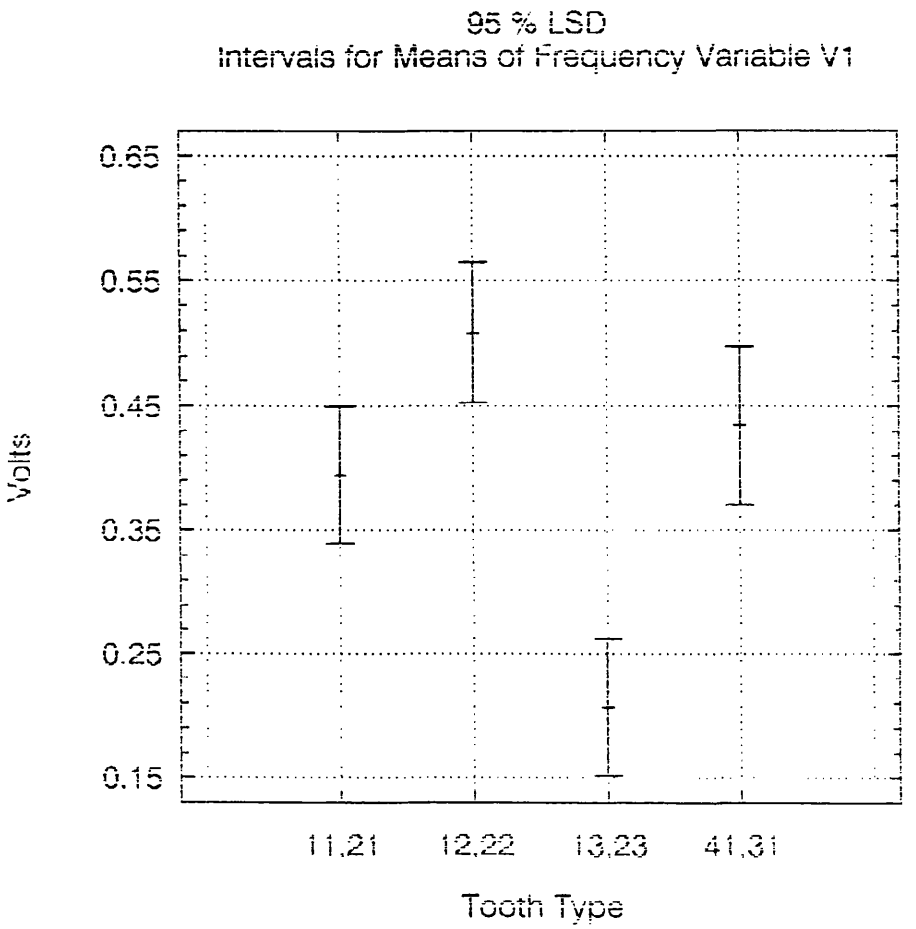


Figure 5.2 (c) 95% least significant difference intervals for flux signal frequency variable V1

Table 5.4 Percentage differences in flux signal variables between 37 pairs of vital anterior teeth (Patients n=10)

Flux signal variable	Mean (SD)	Range (%)
Mean Flux	25.0 (19.4)	0.0-88.0
Mean amplitude of Slow Wave Vasomotion	35.5 (29.1)	0.0-118.0
Mean amplitude of Cardiac Cycle	19.6 (22.0)	0.0-85.7
Frequency variable V1	30.5 (23.7)	0.6-86.7
Frequency variable V2	52.2 (37.2)	0.6-133.3
Frequency variable V3	30.0 (25.4)	2.5-109.0
Frequency variable V4	22.6 (18.9)	1.3-63.7

5.2.4 Discussion

The results showed wide inter-patient and intra-patient ranges for values of all flux signal variables. The large inter-patient variation in the range of Mean Flux values is in agreement with most other published studies. Six studies have reported Mean Flux values from vital permanent anterior teeth and their findings are summarised in Table 5.5. Of these studies, only Wilder-Smith (1988) reported no significant differences in Mean Flux values obtained from different patients. However, the narrow range of Mean Flux values reported by the author are very different from those reported in other studies (Table 5.5) and from the present study (Table 5.1); the difficulties with the recording method used by Wilder-Smith were discussed in Section 4.2.4. Visual analysis of Figure 5.1 (a-g) show that variation of flux signal variables with tooth type follow variation of Mean Flux value. This would be anticipated in view of the significant correlation between Mean Flux and the other flux signal variables demonstrated in Section 3.6.

The wide inter-patient and intra-patient variation in the values of flux signal variables obtained from vital teeth of the same type, under the same recording conditions, were similar to those noted in the study reported in Chapter 4. These wide variations indicate that there would be difficulties in attempting to use laser doppler flowmetry to determine whether a flux signal from a vital tooth differed significantly from a flux signal from a different vital tooth. In addition, the finding that there were significant differences for all flux signal variables, with the exception of amplitude of the Cardiac Cycle, between the four types of anterior teeth indicate that caution should be used when comparing flux signal variables obtained from different tooth types. However, the aim of this part of the study was to determine

Table 5.5. A review of published studies on the range of Mean Flux values obtained from vital teeth using laser doppler flowmetry.

Study	No of Pts	No of teeth	Tooth type	Range of Mean Flux (mean, SD) (arbitrary units)
Olgart *et al.* (1988)	25	33	Incisors	1.6-10.0 (5.18, SD 0.37)
Wilder-Smith (1988)	30	183	All	15.8-18.4 (16.9, SD 7.9)
Vongsavan *et al.* (1991)	8	14	Incisors	2.2-22.7 (7.2, SD 5.0)
Watson *et al.* (1992)	10	17	Incisors	0.6-5.0 (2.0, SD 0.8)
Pitt Ford *et al.* (1993)	10	10	Incisors	8.8-41.4 (15.3, SD 1.8)
Ingolfsson *et al.* (1993)	5	5	Incisors	2.0-5.8 (3.4, SD 1.4)
Present study	10	57	Incisors	7.0-46.0 (20.5, SD 7.8)

whether some flux signals from vital teeth might be incorrectly classified if the L.D.F. classification criteria defined in Section 5.1 were used as the lower limits of normality for flux signals from vital dental pulps.

All Mean Flux values from incisors in the study (n=57) were in excess of 6 P.U., all showed Slow Wave Vasomotion and in 56 of the teeth (98%) the amplitude was in excess of 1.5 P.U. Therefore, if these values for flux signal variables were used to discriminate between vital and non-vital incisor dental pulps, a vital dental pulp would be unlikely to be wrongly classified as non-vital. In addition, 52 out of the 57 incisor pulps showed a regular cardiac pulse and this flux signal variable could be used as an additional diagnostic criterion.

Of the 20 maxillary canine teeth in the study, two teeth, both from different patients, had Mean Flux values of less than 7 P.U. and both had Slow Wave Vasomotion with an amplitude of less than 1.6 P.U.. The L.D.F. classification criteria should, therefore, be applied with caution to maxillary canine teeth as there will be the possibility of a false negative classification for the presence of pulpal blood flow. However, canine teeth are only rarely involved in dental trauma (Section 2.3.1).

5.3 AN INVESTIGATION OF THE REPEATABILITY OF FLUX RECORDINGS FROM ANTERIOR TEETH.

5.3.1 Introduction and Aims

The study reported in Section 5.2 showed that although there was wide variation in flux signal variables obtained from vital anterior teeth, the variation was unlikely to result in vital teeth being classified as non-vital if the diagnostic criteria used were a minimum Mean Flux value of 7 P.U. and a minimum amplitude of Slow Wave Vasomotion of 1.6 P.U.. However, these recordings were made under carefully controlled conditions (see Section 5.2.2). When the pulpal status of traumatised teeth is assessed over a period of time, variation in recording conditions may be

unavoidable; temporal variation may occur and also variation due to eating, exercise, body position and the fabrication of a new impression jig. The aim of this part of the study was to determine if any variation in the recording conditions was likely to result in a vital dental pulp being wrongly classified as necrotic, using the L.D.F. classification criteria outlined in Section 5.1, and to determine the repeatability of flux signals from individual teeth with variation in recording conditions.

5.3.2 Materials and Method

The sample group was described in Section 5.2.2. A typical recording session would involve about 1.5 hours of chairside time and it was, therefore, not practical to include all subjects in all of the studies. However, at least five subjects were always included. The recording method used is described in Appendix A. The recording conditions of the baseline recording session were as described in Section 5.2.2. Variation in these conditions were as follows;

a) Repeating the recording after 10 minutes - Ten minutes after taking a recording, the recording was repeated using the same jig (teeth=19, patients=8).

b) Repeating the recording after 5 weeks - Five weeks after taking a recording, the recording was repeated using the same jig (teeth=28, patients=7).

c) Fabrication of a new impression jig - A new impression jig was constructed and the recordings obtained compared with those obtained using the original jig during the same session (teeth=19, patients=6).

d) Sitting position - A recording was taken with the subject in the normal supine position. Without removing the jig, the back of the chair was then elevated until the subject was seated and the recording repeated (teeth=6, patients=6).

e) Time of day - A recording was taken in the early evening, around 6.0 p.m., and was repeated the following morning at around 8.0 a.m. (teeth=10, patients=5).

f) Exercise - Following a recording the subject ran up five flights of stairs as fast as possible before returning for a repeat recording. The second recording commenced within two minutes of the completion of the exercise (teeth=5, patients=5).

g) Food - The subject fasted for eight hours. A recording was taken, following which the subject had a cooked meal with a hot drink. The subject returned for a repeat recording which commenced within 20 minutes of the completion of the meal (teeth=9, patients=5).

The data in all of the studies in this section consisted of pairs of flux signal recordings, matched for patient and tooth. The differences in flux signal variables between the two recordings following the variation in the recording condition were then analysed. Visual analysis of plots of the differences between paired data against the mean of the data, for all of the variables contained within the flux signal, indicated that they were related; that is as the value of the mean increased, so there was a tendency for the value of the difference to increase. Because of this, the percentage difference between the paired data was analysed, rather than the absolute difference, using Students paired t test. As the seven flux signal variables contained within a flux signal were not completely independent (Section 3.6.4) the level of significance was reduced to $p<0.01$ in order to prevent erroneous rejection of the null hypothesis. The confidence interval and the prediction interval were also calculated. The confidence interval applies to the mean of the sample, whereas the prediction interval applies to an individual and indicates the measurement range within which there would be a 95% chance of a repeat recording of that variable occurring. It is related to the size of the sample, and will generally be at least twice the standard deviation from the mean on each side and larger if the sample size is small. It is calculated by;

$$P.I. = \text{mean} \pm t\sqrt{SD^2\left(1+\tfrac{1}{n}\right)}$$

5.3.3 Results

The results for Mean Flux are shown in Table 5.6, for amplitude Slow Wave Vasomotion in Table 5.7, for amplitude Cardiac Cycle signal in Table 5.8, and for frequency variables V1-V4 in Tables 5.9-5.12. The numbers of recordings satisfying the various criteria for "vitality" (Section 5.1) for all the anterior teeth in the study are shown in Table 5.13 (a) and for incisors only in Table 5.13(b).

Statistical analysis of the results showed no significant change in flux signal variables with change in recording conditions with the exception of a significant increase ($p<0.002$) in Mean Flux produced by the fabrication of a new impression jig. The results show relatively small changes in mean values for flux signal variables with change in recording conditions, but with large standard deviations, and wide 95% confidence and prediction intervals indicating wide variation within individuals. For example, repeating the recording after 10 minutes reduced the mean value of Mean Flux by only 3.2%, but with a standard deviation of 15.1%. Repeating the recording after five weeks increased the mean value of Mean Flux by 5.9%, but with a standard deviation of 43.1%. Exercise increased the mean value of Mean Flux by only 7.8%, but the standard deviation was 54.0% and the prediction interval was between -156.3 and 171.9% of the pre-exercise Mean Flux value. Generally, the best repeatability for all flux signal variables was when the recording was repeated after 10 minutes, and the poorest repeatability was immediately after exercise. There was a tendency for variation in values of amplitude of Slow Wave Vasomotion to be larger than those for Mean Flux and amplitude of the Cardiac Cycle signal, and for variation in frequency variables V2 and V3 (slow wave frequencies) to be larger than for V1 (total of all frequencies) and V4 (cardiac cycle frequencies).

Table 5.6 Mean percentage change in Mean Flux from vital anterior teeth with variation in recording conditions.

Change in recording condition	Mean % change in Mean Flux (SD)	95% Confidence interval	Prediction interval
Repeat after 10 mins. (teeth=19, pts.=8)	-3.8 (15.0)	-11.1 to 3.4	-36.3 to 28.7
Repeat after 5 weeks (teeth=28, pts.=7)	5.9 (43.1)	-10.7 to 22.6	-84.0 to 95.8
New recording jig (teeth=19, pts.=6)	27.4 (32.1)	11.9 to 42.9	-41.9 to 96.7
Patient sitting (teeth=6, pts.=6)	1.4 (16.8)	-16.3 to 19.1	-45.3 to 48.1
Time of day (teeth=10, pts.=5)	13.9 (31.7)	-9.3 to 36.0	-61.2 to 89.0
Post exercise (teeth=5, pts.=5)	7.8 (54.0)	-59.3 to 74.8	-156.3 to 171.9
Post eating (teeth=9, pts.=5)	-5.4 (11.4)	-14.2 to 3.3	-33.0 to 22.2

Table 5.7 Mean percentage change in mean amplitude of Slow Wave Vasomotion from vital anterior teeth with variation in recording conditions.

Change in recording condition	Mean % change in amplitude. SWV (SD)	95% confidence interval	Prediction interval
Repeat after 10 mins. (teeth=19, pts=8)	2.3 (32.8)	-13.5 to 18.1	-68.7 to 73.3
Repeat after 5 weeks (teeth=28, pts.=7)	6.2 (34.8)	-7.2 to 19.7	-66.5 to 78.9
New recording jig (teeth=19, pts.=6)	7.9 (42.9)	-12.7 to 28.6	-61.4 to 77.2
Patient sitting (teeth=6, pts.=6)	13.6 (26.6)	-14.4 to 41.5	-60.3 to 87.5
Time of day (teeth=10, pts.=5)	-35.8 (42.9)	-66.5 to -5.1	-137.6 to 66
Post exercise (teeth=5, pts.=5)	-6.6 (82.0)	-108.4 to 95.2	-255.8 to 242.6
Post eating (teeth=9, pts.=5)	3.5 (19.7)	-11.6 to 18.7	-44.5 to 51.5

Table 5.8 Mean percentage change in amplitude of Cardiac Cycle signal from vital anterior teeth with variation in recording conditions.

Change in recording condition	Mean % change in amplitude Cardiac Cycle (SD)	95% Confidence interval	Prediction interval
Repeat after10 mins (teeth=19, pts.=8)	-6.7 (20.7)	-16.6 to 3.3	-51.5 to 38.1
Repeat after 5 weeks (teeth=28, pts.=7)	3.6 (36.1)	-10.4 to 17.6	-71.8 to 79
New recording jig (teeth=19, pts.=6)	11.7 (21.1)	1.5 to 21.9	-33.8 to 57.2
Patient sitting (teeth=6, pts.=6)	-13.3 (20.7)	-35.0 to 8.4	-70.7 to 44.1
Time of day (teeth=10, pts.=5)	19.4 (30.7)	-2.5 to 41.4	-53.3 to 92.1
Post exercise (teeth=5, pts.=5)	9.1 (46.2)	-48.2 to 66.5	-131.3 to 149.5
Post food (teeth=9, pts.=5)	9.6 (15.5)	-2.2 to 21.5	-28 to 47.2

Table 5.9. Mean percentage change in flux signal frequency variable V1 from vital anterior teeth with variation in recording conditions.

Change in recording condition	Mean % change in V1 (SD)	95% Confidence interval	Prediction interval
Repeat after10 mins (teeth=19, pts.=8)	1.1 (21.1)	-9.1 to 11.3	-44.3 to 46.5
Repeat after 5 weeks (teeth=28, pts.=7)	-1.4 (30.0)	-13.1 to 10.2	-64.1 to 61.3
New recording jig (teeth=19, pts.=6)	12.1 (35.6)	-5.0 to 29.3	-66.5 to 90.7
Patient sitting (teeth=6, pts.=6)	-5.9 (18.6)	-25.5 to 13.6	-57.5 to 45.7
Time of day (teeth=10, pts.=5)	-7.9 (36.4)	-33.9 to 18.2	-94.2 to 78.4
Post exercise (teeth=5, pts.=5)	-1.5 (76.9)	-97.1 to 94.1	-235.4 to 232.4
Post eating (teeth=9, pts.=5)	-23.6 (37.4)	-52.3 to 5.2	-114.4 to 67.3

Table 5.10 Mean percentage change in flux signal frequency variable V2 from vital anterior teeth with variation in recording conditions.

Change in recording condition	Mean % change in V2 (SD)	95% Confidence interval	Prediction interval
Repeat after10 mins (teeth=19, pts.=8)	2.3 (40.4)	-17.2 to 21.8	-84.8 to 89.4
Repeat after 5 weeks (teeth=28, pts.=7)	9.4 (51.6)	-10.7 to 29.4	-98.3 to 117.1
New recording jig (teeth=19, pts.=6)	15.8 (59.3)	-12.8 to 44.4	-112.0 to 143.6
Patient sitting (teeth=6, pts.=6)	-14.2 (51.2)	-67.9 to 39.6	-156.5 to 128.1
Time of day (teeth=10, pts.=5)	-20.4 (55.8)	-60.4 to 19.5	-152.9 to 112.1
Post exercise (teeth=5, pts.=5)	-22.3 (88.1)	-131.7 to 87.2	-290.2 to 245.6
Post eating (teeth=9, pts.=5)	-39.0 (53.5)	-80.2 to 2.1	-169.0 to 91.0

Table 5.11 Mean percentage change in flux signal frequency variable V3 from vital anterior teeth with variation in recording conditions.

Change in recording conditions	Mean % change in V3 (SD)	95 % Confidence interval	Prediction interval
Repeat after10 mins (teeth=19, pts.=8)	7.4 (31.7)	-7.9 to 22.6	-60.9 to 75.7
Repeat after 5 weeks (teeth=28, pts.=7)	-8.1 (39.3)	-23.3 to 7.2	-90.1 to 73.9
New recording jig (teeth=19, pts.=6)	8.8 (39.3)	-10.2 to 27.7	-75.9 to 93.5
Patient sitting (teeth=6, pts.=6)	0.0 (18.7)	-20.6 to 18.7	-52.0 to 52.0
Time of day (teeth=10, pts.=5)	-12.2 (42.5)	-42.6 to 18.1	-112.9 to 88.5
Post exercise (teeth=5, pts.=5)	12.5 (64.2)	-67.3 to 92.3	-182.9 to 207.9
Post eating (teeth=9, pts.=5)	-24.7 (46.8)	-60.6 to 11.3	-138.5 to 89.1

Table 5.12 Mean percentage change in flux signal frequency variable V4 from vital anterior teeth with variation in recording conditions.

Change in recording condition.	Mean % change in V4 (SD)	95% Confidence interval	Prediction interval
Repeat after10 mins (teeth=19, pts.=8)	-7.8 (17.4)	-16.2 to 0.6	-45.3 to 29.7
Repeat after 5 weeks (teeth=28, pts.=7)	1.1 (30.1)	-10.6 to 12.7	-61.6 to 63.8
New recording jig (teeth=19, pts.=6)	8.8 (21.7)	-1.7 to 19.2	-37.9 to 55.5
Patient sitting (teeth=6, pts.=6)	-2.3 (15.0)	-18.0 to 13.5	-43.9 to 39.3
Time of day (teeth=10, pts.=5)	21.9 (23.9)	4.8 to 39.0	-34.7 to 78.5
Post exercise (teeth=5, pts.=5)	12.0 (43.9)	-42.4 to 66.5	-121.4 to 145.4
Post eating (teeth=9, pts.=5)	-6.5 (12.0)	-15.7 to 2.7	-35.6 to 22.6

Table 5.13 (a) Presence of Mean Flux ≥ 7 Perfusion Units (P.U.), Slow Wave Vasomotion (SWV) with an amplitude ≥ 1.6 P.U. and a regular cardiac pulse signal in the flux signal from vital anterior teeth (incisors and canines) with changes in recording conditions.

Study	Number of recordings with Mean Flux ≥7 P.U.	Number of recordings with amplitude S.W.V ≥1. 6 P.U.	Number of recordings with a regular cardiac pulse
Baseline (teeth=77, pts.=10)	73 (95%)	72 (94%)	66 (86%)
Repeat after 10 mins (teeth=19, pts.=8)	19 (100%)	16 (84%)	15 (79%)
Repeat after 5 weeks (teeth=28, pts.=7)	28 (100%)	26 (93%)	23 (82%)
New recording jig (teeth=19, pts.=6)	19 (100%)	19 (100%)	16 (84%)
Patient sitting (teeth=6, pts.=6)	6 (100%)	6 (100%)	4 (teeth=5) (80%)
Time of day (teeth=10, pts.=5)	10 (100%)	10 (100%)	10 (100%)
Post Exercise (teeth=5, pts.=5)	5 (100%)	4 (80%)	3 (teeth=4) (75%)
Post eating (teeth=9, pts.=5)	9 (100%)	8 (89%)	8 (89%)

Table 5.13 (b) Presence of Mean Flux ≥ 7 Perfusion Units (P.U.), Slow Wave Vasomotion (SWV) with an amplitude ≥ 1.6 P.U. and a regular cardiac pulse signal in the flux signal from vital anterior teeth (incisors only) with changes in recording conditions.

Study	Number of recordings with Mean Flux ≥7 P.U.	Number of recordings with amplitude S.W.V ≥1. 6 P.U.	Number of recordings with a regular cardiac pulse
Baseline (teeth=57, pts.=10)	57 (100%)	56 (98%)	52 (91%)
Repeat after 10 mins (teeth=13, pts.=8)	13 (100%)	13 (100%)	9 (69%)
Repeat after 5 weeks (teeth=20, pts.=7)	20 (100%)	20 (100%)	19 (95%)
New recording jig (teeth=14, pts.=6)	14 (100%)	14 (100%)	13 (93%)
Patient sitting (teeth=6, pts.=6)	6 (100%)	6 (100%)	4 (teeth=5) (80%)
Time of day (teeth=10, pts.=5)	10 (100%)	10 (100%)	10 (100%)
Post Exercise (teeth=5, pts.=5)	5 (100%)	4 (80%)	3 (teeth=4) (75%)
Post eating (teeth=9, pts.=5)	9 (100%)	8 (89%)	8 (89%)

5.3.4 Discussion

The results indicate poor repeatability of all flux variables contained within a flux signal. Even repeating the recording after 10 minutes which, as might be anticipated, gave the best repeatability of all the changes in recording conditions tested, produced a prediction interval of -35.4 to 29.0% for Mean Flux, with all the remaining variables generally having even wider prediction intervals. It is of interest that the flux signal variables with the poorest repeatability were those associated with Slow Wave Vasomotion, that is amplitude of Slow Wave Vasomotion and flux frequency variables V2 and V3. Figure 3.10, showing a continuous recording of pulpal blood flow over 30 minutes, demonstrates the inherent variability in Slow Wave Vasomotion.

The mean percentage change in Mean Flux values over five weeks of 5.9% (28 teeth, 7 subjects) compares with a mean percentage change of 4.8% (8 teeth, 8 subjects) reported by Aars *et al.* (1993) and a mean percentage change of -7.1% (5 teeth, 5 subjects) reported by Gazelius *et al.*(1986). However, the relatively small mean percentage change in Mean Flux of 5.9% in the present study had a standard deviation of 43.1%, indicating poor repeatability of individual flux signal recordings.

A surprising finding was the significant increase ($p<0.002$) in Mean Flux produced by fabrication of a new impression jig. Gazelius *et al.* (1988) reported that this procedure did not result in a significant change in Mean Flux value. One possible reason for the significant increase in Mean Flux in the present study was that all the new impression jigs had the probe holes prepared at the same session. An initial operator error in assessing probe angulation (see Section 3.5.2, and Figure 3.9) was, therefore, likely to be repeated on subsequent jigs.

In the present study, exercise did not cause a significant change in the mean value of any flux signal variable, although it did have the greatest effect on the repeatability of flux signal variables of all factors investigated. Aars *et al.* (1992) reported that exercise increased pulpal blood flow in all eight subjects tested, with the mean increase in flux being about 15%. However, Watson *et al.* (1992) reported a

variation in response in 10 subjects, with pulpal blood flow increasing in some and decreasing in others, with a mean percentage change of 38% from resting level.

For some of the studies in Section 5.3 more than one tooth per patient were used: the range was from one tooth per patient for the exercise and sitting study to a maximum of six teeth per patient for the study involving fabrication of a new impression jig. For each tooth the baseline value acted as an intra-tooth control value for the subsequent study, allowing a matched paired statistical analysis.

Of the 96 flux signal recordings taken following application of a recording variable, all had a Mean Flux value ≥ 7 P.U.. Two flux signals had an amplitude of Slow Wave Vasomotion of <1.6 P.U.; one was post exercise and one was post eating. However, both recordings were from the same subject, who also provided the only incisor tooth from the sample of 57 baseline recordings which had an amplitude of Slow Wave Vasomotion <1.6 P.U. Other flux signal variables which may indicate the correct classification of such a signal are discussed in Section 6.5.2. In general it would appear that although changing the recording conditions reduces the repeatability of flux signal recordings, the variation is unlikely to affect the reliability of laser doppler flowmetry in determining the presence or absence of pulpal blood flow.

5.3.5 Summary and conclusion

The poor repeatability of flux signal recordings means that it is unlikely that laser doppler flowmetry will allow the reliable quantification of change in pulpal blood flow in vital teeth over time. However, the main aim of this chapter was to assess the reliability of the L.D.F. classification criteria described in Section 5.1 in correctly classifying vital dental pulps. All 134 recordings from 57 vital incisor teeth from 10 patients, recorded under a variety of different conditions, had Mean Flux values ≥ 7 P.U.. In addition, 131 (98%) had an amplitude of Slow Wave Vasomotion ≥ 1·6 P.U.. It would seem, therefore, that these L.D.F. classification criteria were unlikely to result in a vital pulp being classified as non-vital. In Chapter 6 these L.D.F.

classification criteria will be applied to a sample of non-vital teeth to determine their reliability in correctly classifying non-vital dental pulps.

CHAPTER 6

LASER DOPPLER FLOWMETRY OF THE NON-VITAL DENTAL PULP.

6.1 INTRODUCTION AND AIMS

The study reported in Chapter 5 demonstrated that laser doppler flowmetry (L.D.F) of vital dental pulps produced flux signals with Mean Flux values ≥ 7 P.U. and with Amplitude of Slow Wave Vasomotion ≥ 1.6 P.U., these values being referred to as the L.D.F. classification criteria. In this chapter the flux signals recorded from non-vital dental pulps will be investigated with the following aims:

a) to determine the values of flux signals from non-vital dental pulps.

b) to determine the validity of the L.D.F. classification criteria for discriminating between the flux signals from non-vital and vital dental pulps.

c) to correlate the laser doppler flowmetry classification with clinical and histopathological findings subsequent to pulpectomy.

d) to correlate the laser doppler flowmetry classification with other methods of assessing pulpal status.

6.2 MATERIALS AND METHOD

The study sample comprised patients attending the Department of Child Dental Health, Glasgow Dental Hospital and School, for review following dental trauma. Data were collected from 67 non-vital anterior teeth (55 patients, mean age 13.5 years, range 8.0-33.5 years) for which pulpectomy was planned. The criteria for pulpectomy were in line with current guidelines, being a diagnosis of irreversible pulpal necrosis based on at least two clinical signs, usually loss of pulpal sensibility and one or more other signs such as radiographic change or coronal discolouration (Andreasen, 1988). For comparison, data were also collected from 84 vital anterior teeth from some of the same and some other patients attending the trauma review clinic (84 patients, mean age 12.4 years, range 6.5-33.5 years). Of the 55 patients

with non-vital teeth, 46 patients (84% of the sample) had an untraumatised anterior tooth available in the same dental arch as the non-vital teeth and these patients were included in the sample of patients with vital teeth. Patients with non-vital teeth where an untraumatised control tooth was unavailable were included in the study as use of the L.D.F. classification criteria outlined in Section 6.1 allowed classification of a laser doppler flowmetry signal independently of an intra-patient control (see Section 6.3.2 for discussion).

A full history and examination of the patient was carried out in accordance with normal clinical practice. The tooth referred for pulpectomy was examined as was an untraumatised anterior tooth from the same arch, if available. The diagnostic tests used to assess pulpal status were as follows:

Tests involving history and examination;

- symptoms of pain
- tenderness of the alveolus to palpation
- alveolar sinus
- increased mobility in an axial direction
- tenderness to percussion
- coronal discolouration to direct light
- coronal discolouration on transillumination

Special investigations

- tooth sensibility to thermal stimulation with ethyl chloride
- tooth sensibility to electric pulp testing
- radiographic examination
- laser doppler flowmetry

The tests of symptoms of pain, alveolar tenderness and tenderness to percussion were not used for the first two weeks, and the test of mobility for the first four weeks,

following traumatic injury. The Analytic Pulp Tester (Analytical Technology, Redmond, Washington 98052) was used for electric pulp testing, with the electrode applied to the incisal edge of the tooth being tested (Section 2.7.5). The dental pulp was classified as non-vital if there was no response or only a weak response at the highest output of the pulp tester. The radiographic criteria assessed were periapical radiolucency, apical root resorption and inflammatory external root resorption. An increase in the width of the periodontal ligament around the periapical region of the study tooth > 0.2 mm was recorded as a periapical lesion. This feature was not recorded as a diagnostic criterion if the tooth had sustained a luxation injury within the previous three months (Andreasen, 1986). To minimise patient X ray exposure, a radiograph was not normally taken if a radiograph had been taken in the previous few weeks and the evidence that the tooth required a pulpectomy was overwhelming (discharging sinus, alveolar swelling, pain). Radiographs were not assessed for the study if taken more than two weeks from the time of pulpectomy. The radiograph of choice was an orthoradial periapical view using a film holder and a long cone paralleling technique. Films were examined by the author in a darkened room, using a backlit X ray viewer and a 3x magnifying lens. Measurements of any periapical lesions noted, and of the diameter of the apical foramen, were made using a travelling microscope with customised optics giving a magnification of 2x. All measurements were made twice, and the average value noted. Laser doppler flowmetry recordings were taken from teeth using the method described in Appendix A. During the course of the study, computer software was developed to carry out frequency analysis of the flux signal. After the software became available the flux signal was stored on an IBM compatible PC (Section 3.6).

The pulpectomy was commenced without local anaesthesia. Rubber dam was applied and access gained to the pulp chamber using a high speed handpiece. A sterile rosehead bur in a slow handpiece was then used to enlarge the access cavity and a

barbed broach introduced into the root canal to remove any pulpal contents. Pulp chamber contents were classified clinically as follows:

a) vital - perfused, bleeding tissue was found immediately on entering the pulp chamber.

b) autolysed - the root canal contents were found to have undergone autolysis. The root canal was either empty or contained pus.

c) necrotic - the pulp chamber and root canal were found to contain non-sensitive, non-perfused necrotic pulp tissue which was removed with a barbed broach.

d) necrosis confined to coronal pulp - the pulp chamber was found to contain non-sensitive, non-perfused necrotic pulp tissue, but the root canal contained some perfused tissue.

Pulpal tissue retrieved following pulpectomy was fixed in 10% buffered formal saline. Tissue specimens were then embedded in paraffin wax and sectioned longitudinally into ribbon sections at levels through the block ensuring adequate sampling. Sections were stained with haematoxylin and eosin stain. Pulps were classified by their histological appearance according to the following criteria:

a) vital - cellular appearance was of normal pulp tissue throughout the specimen, with no signs of nuclear degeneration.

b) partial necrosis - areas of specimen showed signs of necrosis, with loss of cellular outline and nuclear degeneration, while other areas had a normal appearance.

c) total necrosis - the entire specimen appeared necrotic, with loss of cellular outline and nuclear degeneration.

The findings from the diagnostic tests and the pulpectomy were recorded in the clinical notes and then entered into a customised data base in an IBM compatible PC.

The results will be reported and discussed in four sections. These will comprise an analysis of the flux signals obtained (Section 6.3), the clinical findings following

pulpectomy (Section 6.4), the histopathology of the extirpated dental pulps (Section 6.5) and an analysis of the results obtained from the other diagnostic methods reported in 6.2. (Section 6.6).

6.3 ANALYSIS OF FLUX SIGNALS FROM NON-VITAL AND VITAL TEETH.

6.3.1 Results

Laser doppler flowmetry recordings were obtained from 67 non-vital teeth where the pulpal diagnosis could be confirmed by pulpectomy, and from 84 vital teeth. All 67 non-vital teeth subjected to pulpectomy were found clinically to have pulpal necrosis affecting at least the coronal pulp (Section 6.4). Mean Flux values and Amplitude of the Cardiac Cycle values were available for all the recordings. For seven recordings from vital teeth (8% of the sample) data on the flux signal variable Amplitude of Slow Wave Vasomotion were not included in the data subject to statistical analysis. This was due to adverse recording conditions (usually patient restlessness) limiting the recording time, so that three sequential cycles of Slow Wave Vasomotion were not available for analysis (Section 3.5.3). However, at least two cycles of Slow Wave Vasomotion were available for these recordings and these were, in all cases, in excess of the L.D.F. classification criteria for this flux signal variable. Data on the regularity of the Cardiac Pulse signal were available for 42 of the non-vital teeth (74% of the sample) and for 60 of the vital teeth (71% of the sample). Some data for this flux signal variable were not available due to the occasional absence of assistance to alter the chart speed of the pen recorder (Appendix A). Flux signal frequency analysis was available for 24 non-vital teeth (36% of the sample) and 24 vital teeth (29% of the sample) as the computer software became available during the later stages of the study.

Fifty seven (85%) of the 67 non-vital teeth subject to pulpectomy had flux signal recordings where the Mean Flux value fell below the L.D.F. classification

criteria. These recordings were classified as L.D.F. Non-vital. For the remaining 10 non-vital teeth the Amplitude of Slow Wave Vasomotion was below the criteria but Mean Flux values were in excess; these recordings were classified as L.D.F. Intermediate vitality. All 84 vital teeth had flux signal variables in excess of the L.D.F. classification criteria and were classified as L.D.F. Vital. Examples of the three L.D.F. classification categories are shown in Figure 6.1.

The mean values of Mean Flux for the 67 non-vital teeth and the 84 vital teeth are shown in Table 6.1 and in Figure 6.2. The values for Amplitude of Slow Wave Vasomotion are shown in Table 6.2 and Figure 6.3, and for the Cardiac Cycle in Table 6.3 and Figure 6.4. The mean values of the frequency variables are shown in Table 6.4, and Figure 6.5 shows the values for variable V1.

Statistical analysis of the flux signal variables Mean Flux, Amplitude of Slow Wave Vasomotion, Amplitude of Cardiac Cycle and flux signal frequency variables V1, V2, V3 and V4 using one-way analysis of variance demonstrated significant differences ($p<0.0001$) between the three L.D.F. classification groupings of Non-vital, Intermediate vitality and Vital. Further analysis using least significant difference multiple range tests demonstrated no significant difference in the values of the flux signal variables between the L.D.F. groupings Non-vital and Intermediate vitality, but that both groupings were significantly different from the L.D.F. Vital grouping ($p<0.0002$ for frequency variable V2, otherwise $p<0.0001$).

Eight teeth (in eight patients) referred for pulpectomy due to clinical signs of irreversible pulpal necrosis were found to have flux signals with values in excess of the L.D.F. classification criteria. It was felt to be unethical to subject these teeth to pulpectomy as an unnecessary pulpectomy is certainly to the patient's disadvantage (Section 2.4.2), and they were reviewed. Two teeth, both maxillary central incisors, were associated with a discharging labial alveolar sinus. Further clinical investigations

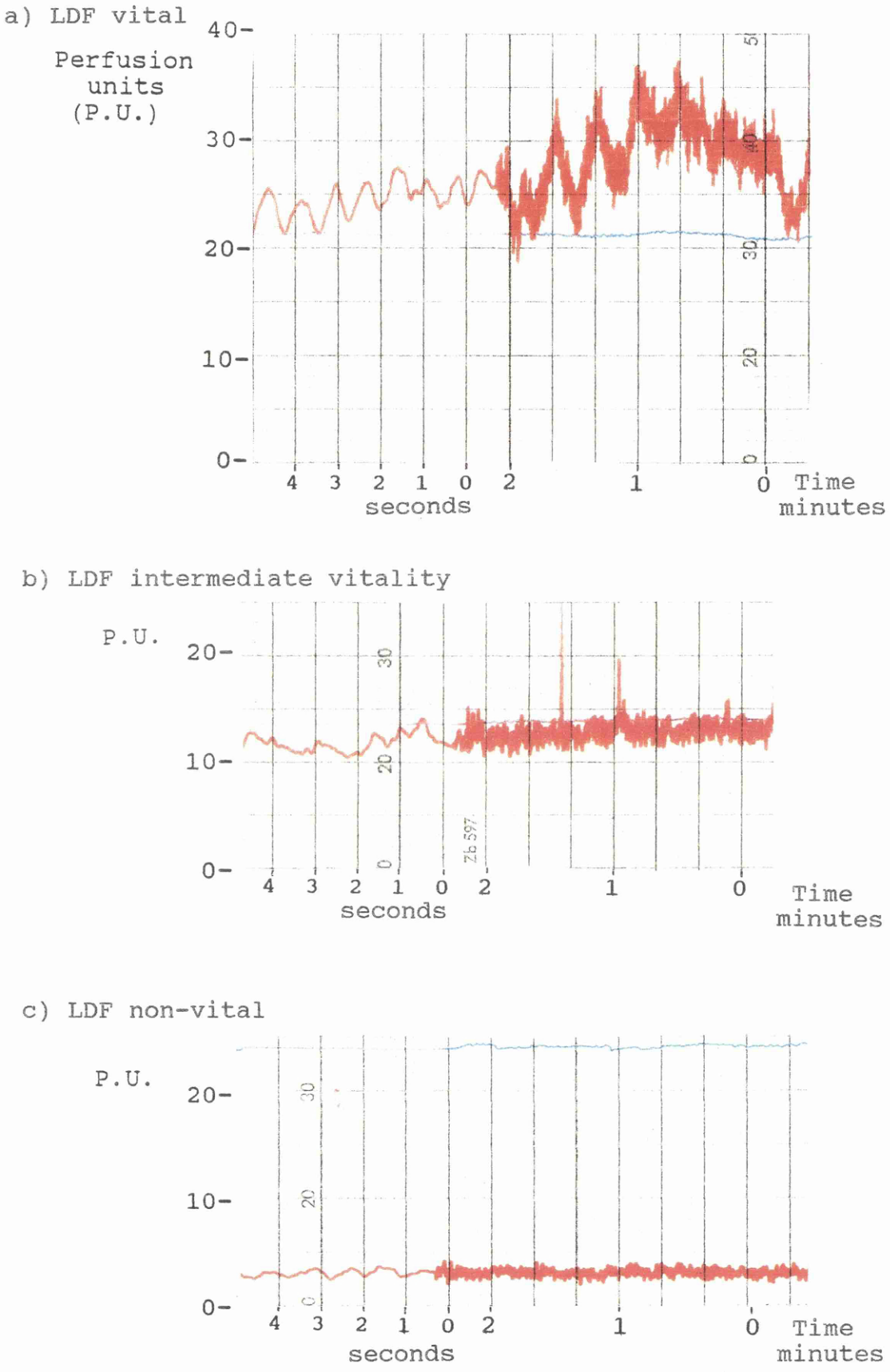


Figure 6.1 Examples of flux signals classified as LDF Vital (a), LDF intermediate vitality (b) and LDF non-vital (c).

Table 6.1 Mean flux values with L.D.F. classification of pulpal status (n=sample size)

LDF classification	Mean value of Mean Flux (P.U.) (SD)	Range of Mean Flux (P.U.)	Mean of Control/pulpectomy Mean Flux ratio	Standard deviation and range of signal ratio
LDF Non-vital (n=57)	2.6 (1.6)	0-6	0.13 (n=53)	S.D. 0.08 0.01-0.33
LDF Intermediate vitality (n=10)	8.5 (2.0)	7-13	0.38	S.D. 0.12 0.27-0.53
LDF Vital (n=84)	25.6 (13.4)	8-82	*	*

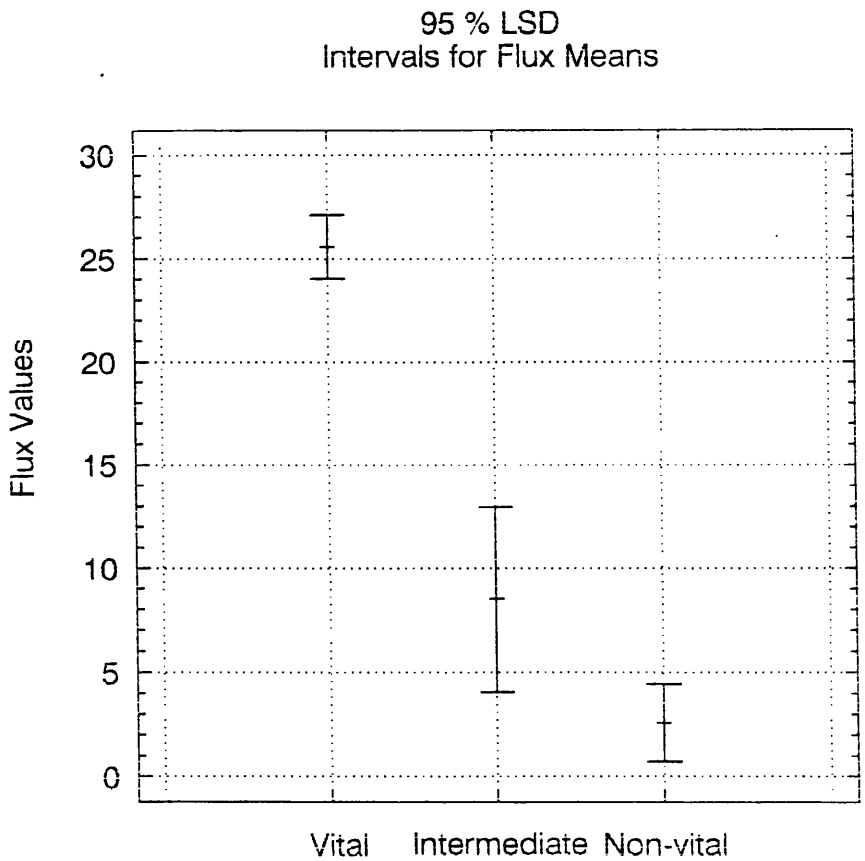


Figure 6.2 95% least significant difference intervals for Mean Flux with L.D.F. classification of pulpal status

Table 6.2 Amplitude of Slow Wave Vasomotion (SWV) with L.D.F. classification of pulpal status (n=sample size)

LDF classification	Mean amplitude of SWV (P.U.) (S.D.)	Range of SWV (P.U.)	Percentage with amplitude SWV ≥1.6 P.U
LDF Non-vital (n=57)	1.0 (0.1)	1.0-1.7	2%
LDF Intermediate vitality (n=10)	1.1 (0.1)	1.0-1.3	0%
LDF Vital (n=77)	4.6 (2.3)	1.6-11.8	100%

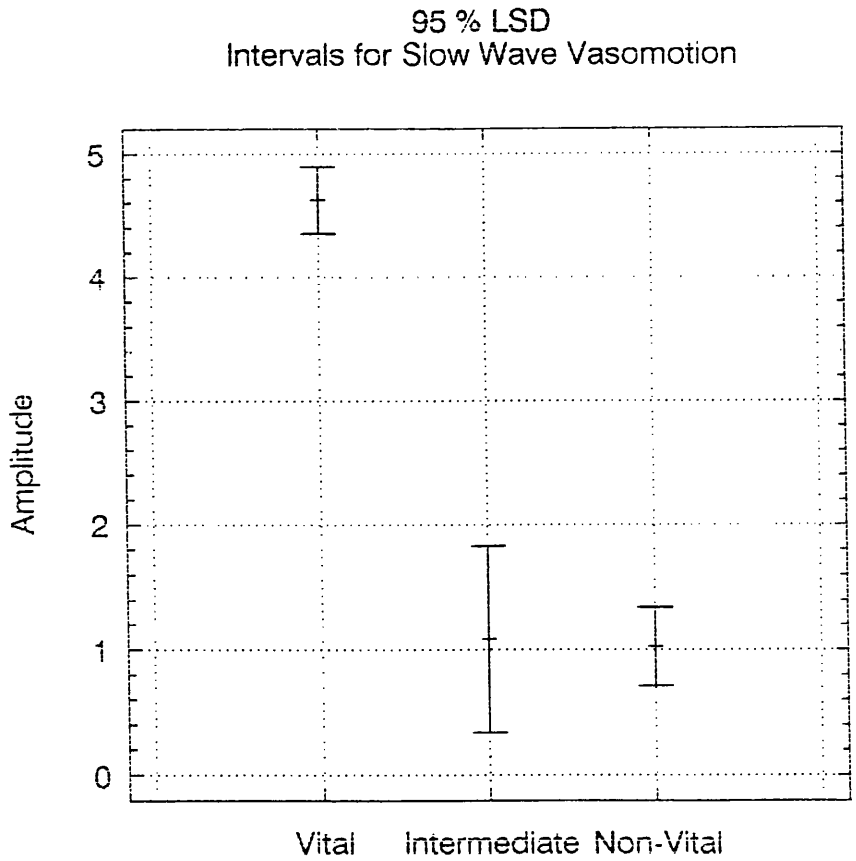


Figure 6.3 95% least significant difference intervals for Amplitude of Slow Wave Vasomotion with L.D.F. classification of pulpal status

Table 6.3 Cardiac Cycle signal values with L.D.F. classification of pulpal status (n=sample size)

LDF classification	Mean amplitude cardiac pulse (P.U.) (SD)	Percentage with cardiac pulse amplitude >2 P.U	Percentage with regular cardiac pulse
LDF Non-vital (n=57)	1.8 (0.4)	0%	0% (n=33)
LDF Intermediate Vitality (n=10)	2.2 (0.4)	20%	33% (n=9)
LDF Vital (n=84)	5.0 (2.1)	98%	95% (n=60)

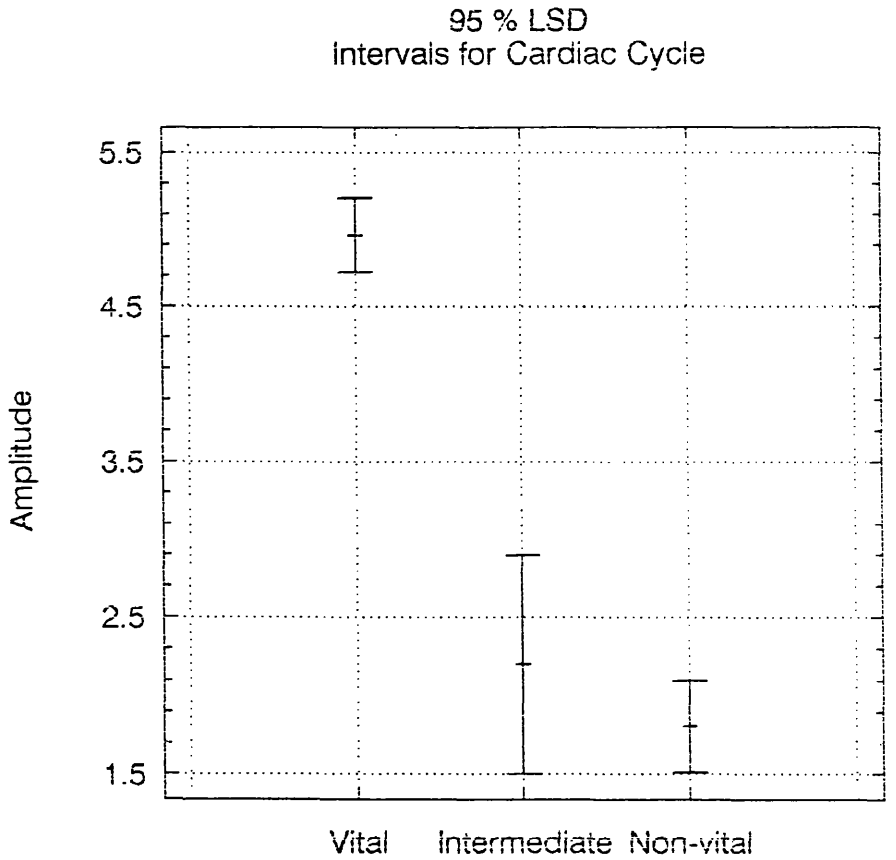


Figure 6.4 95% least significant difference intervals for Amplitude of Cardiac Cycle with L.D.F. classification of pulpal status

Table 6.4 Flux signal frequency variables with L.D.F. classification of pulpal status (n=sample size)

LDF classification	V1 (volts) (SD) Range	V2 (volts) (SD) Range	V3 (volts) (SD) Range	V4 (volts) (SD) Range
LDF Non-vital (n=18)	0.12 (0.03) 0.08-0.18	0.04 (0.03) 0.01-0.13	0.06 (0.03) 0.03-0.12	0.06 (0.01) 0.04-0.08
LDF Intermediate Vitality (n=6)	0.14 (0.07) 0.08-0.24	0.07 (0.05) 0.02-0.13	0.08 (0.04) 0.04-0.13	0.07 (0.02) 0.05-0.11
LDF Vital (n=24)	0.45 (0.24) 0.19-1.32	0.24 (0.20) 0.06-0.97	0.28 (0.15) 0.10-0.80	0.17 (0.09) 0.09-0.43

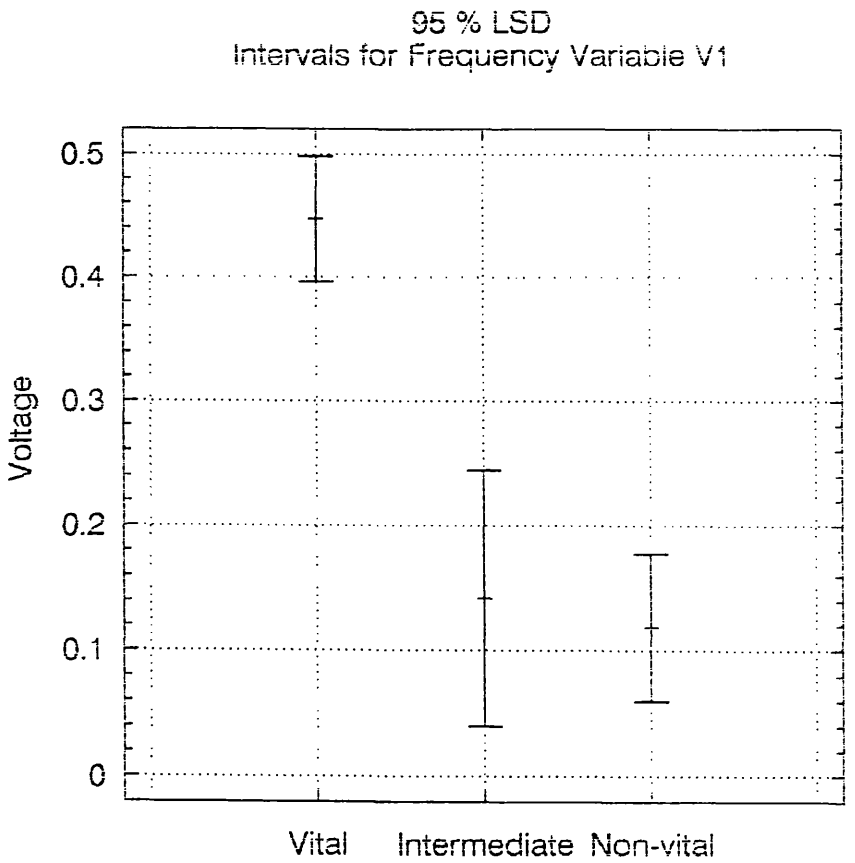


Figure 6.5 95% least significant difference intervals for flux signal frequency variable V1 with L.D.F. classification of pulpal status

revealed that for one patient the sinus was due to retained fragments of a traumatised primary incisor, with the sinus resolving following curettage. The sinus in the second patient was found to be due to mucous secreting glandular tissue having been incorporated into the repair of a maxillary cleft. This sinus resolved following surgical curettage of the area. Of the remaining six teeth, four teeth demonstrated continued root growth radiographically from the time of the L.D.F. Vital classification, although one of the teeth was classified L.D.F. Intermediate vitality 10 months following the initial L.D.F. assessment. This tooth, which had sustained a crown fracture, is still under review. Of the remaining two teeth, one tooth was classified L.D.F. Intermediate vitality seven months from the initial L.D.F. Vital assessment and then L.D.F. Non-vital one year following the initial assessment, with the L.D.F. classification confirmed by pulpectomy. This tooth had sustained a crown fracture at the time of trauma. The final tooth was classified L.D.F. Vital four years after trauma, and showed radiographic evidence of continued root development between the time of trauma and the L.D.F. Vital assessment. Reviewed without laser doppler flowmetry nine months after the initial L.D.F. classification, the tooth showed restored sensibility to electric pulp testing and no radiographic signs of pathology. However, the patient presented with pain from the tooth two and a half years after the initial L.D.F. assessment (six and a half years following the trauma) and the tooth was found to be non-vital at pulpectomy. This tooth had sustained an extensive crown fracture at the time of trauma.

6.3.2 Discussion

The reliability of a diagnostic test can be expressed in terms of its sensitivity and specificity. The sensitivity of a dental pulp diagnostic test is a measure of its ability to identify non-vital pulps. It is determined by the number of true positives (for pulpal necrosis) divided by the number of true positives plus false negatives, and is expressed as a fraction of 1.0, or more usually as a decimal. For example, a pulpal diagnostic

test with a sensitivity of 0.9 will detect 90 % of non-vital pulps contained within a sample. The specificity of a test is a measure of its ability to identify vital pulps and is determined by the number of true negatives (for pulpal necrosis) divided by the number of true negatives plus the false positives. A pulpal diagnostic test with a specificity of 0.9 will correctly identify 90% of vital pulps in a sample. It will, however, incorrectly classify the remaining 10% of vital pulps as being non-vital. The ideal diagnostic test should, therefore, have a sensitivity of 1.0 and a specificity of 1.0. The relative importance of these two factors, which are often inversely related, depends on the disease that is being diagnosed. If a disease has serious consequences for a patient and the treatment has no complications for the patient who does not have the disease, then diagnostic tests of the highest sensitivity should be used, if necessary at the expense of a loss of specificity. However, pulpal necrosis does not generally have serious implications for the patient and delay until the diagnosis is clear need not have a significant effect on the prognosis of treatment (Section 2.4.2). On the other hand, there are obvious disadvantages associated with the unnecessary removal of a vital pulp from a tooth, especially if root formation is incomplete (Cvek, 1992). Therefore, for pulpal diagnostic tests, a balance of high specificity against lower sensitivity is usually in a patient's interest.

The sensitivity and specificity of laser doppler flowmetry as a pulpal diagnostic aid is dependent on the method used to classify the flux signal. In the present study flux signals were classified using the L.D.F. classification criteria outlined in Section 6.1. Flux signal variables Mean Flux and Amplitude of Slow Wave Vasomotion were used as absolute measurements, interpreted with reference to population derived mean normal values, so that intra-patient controls were unnecessary. This is a similar method to that used, for example, when assessing periodontal health using the Community Periodontal Index of Treatment Need (Ainamo, Barnes & Beagrie, 1982), when assessing dental health using the Decayed, Missing and Filled Index (Todd & Dodd, 1985) or when assessing incisor overjet as part of the Index of

Orthodontic Treatment Need (O'Brien, 1994). In the present study an untraumatised intra-patient control tooth was not always available since dental trauma commonly affects more than one tooth in a mouth (see Section 2.3.1).

The results indicate that the L.D.F. classification criteria outlined in Section 6.1 formed a reliable basis for discriminating between flux signals from non-vital and vital dental pulps, with a sensitivity and specificity of 1.0. The majority of flux signals from the 67 non-vital teeth (83% of the sample) had Mean Flux and Amplitude of Slow Wave Vasomotion values less than the L.D.F. classification criteria while for all the 84 vital teeth, the values for the same flux signal variables were in excess of both of these criteria. Where only one L.D.F. classification criterion was exceeded (as in the L.D.F. Intermediate vitality group), the tooth was still found clinically to be non-vital. In view of this, and as no significant difference in flux signal variables between the L.D.F. Intermediate and L.D.F. Non-vital teeth was found, both groups can be considered together as L.D.F. Non-vital. However, although the appropriate L.D.F. classification was clearly indicated in the majority of teeth tested (Figures 6.2-6.5), the range of values for the individual flux signal variables included in the L.D.F. classification criteria showed some overlap between the classification groupings (Table 6.1 and 6.2). This resulted in a few teeth tested having flux signal variables relatively close to the values of the L.D.F. classification criteria. It would be expected that in a larger sample there would be an increase in the range of values for flux signal variables, with the consequence that a very few teeth may be incorrectly classified. An additional factor for consideration is that the measurement error in quantifying flux signal variables (Section 3.5) will become an increasingly important factor affecting the reliability of the L.D.F. classification criteria as the values of the subject tooth flux signal variables approach those of the classification criteria. Other flux signal variables should, therefore, be considered when classifying a 'borderline" recording. For example, one flux signal from 57 signals from a different sample of non-traumatised anterior teeth described in

Chapter 5 was classified as L.D.F. Intermediate vitality due to an Amplitude of Slow Wave Vasomotion of 1.3 P.U. This would give a specificity for the L.D.F. classification criteria of 0.98. However, the Amplitude of the Cardiac Cycle was greater than 2 P.U. and it contained a regular cardiac pulse. Reference to Table 6.3 shows that these are flux signal characteristics most commonly associated with L.D.F. Vital rather than L.D.F. Intermediate Vitality signals.

The ranges of the flux signal frequency variables V1-V4 also showed minor overlap between the three L.D.F. classification groupings, indicating that they were not entirely discreet. However, these flux signal variables were computed, thus avoiding the measurement error associated with quantifying flux signal variables visually from chart recordings (Section 3.5). In addition, they were also readily available at the chairside whereas quantifying the chart recordings involved leaving the clinical area to carry out the required calculations. With a larger sample size, these flux signal variables could form the basis of a computer-aided expert diagnostic system.

The decision to defer pulpectomy on the eight teeth which were referred for pulpectomy but were found to be L.D.F. Vital would seem to have been justified. Five of the teeth went on to show signs of continued dental pulp vitality; for two teeth the alveolar sinus resolved following curettage while the other three teeth demonstrated continued root growth on subsequent radiographs. It is of interest that the three teeth from the group which became non-vital had all sustained coronal fractures at the time of trauma. The reduction in the prognosis for pulpal healing following dental trauma if a crown fracture had been sustained was discussed in Section 2.3.5.

6.4 CLINICAL FINDINGS FOLLOWING PULPECTOMY

6.4.1 Results

Pulpectomies were carried out on a total of 67 non-vital permanent anterior teeth. In all cases the cause of the suspected dental pathology was trauma. The sample included one maxillary canine, 12 maxillary lateral incisors, 49 maxillary central incisors and five mandibular incisors. The clinical findings are grouped according to the L.D.F. classification of L.D.F. Intermediate vitality (n=10) and L.D.F. Non-vital (n=57), and are presented in Table 6.5.

Ten pulpectomies were carried out where the L.D.F. classification was Intermediate Vitality. In all cases access to each pulp chamber was gained without local anaesthesia and without any of the patients reporting pain. In all cases the contents of the pulp chamber were found to be necrotic and non-perfused. Three cases were found to have perfused tissue in the apical half of the canal. For two of the cases this resulted in only minor haemorrhage and discomfort during root canal instrumentation, which was adequately controlled by irrigating the root canal with local anaesthetic solution. For one of the cases the haemorrhage was particularly profuse and clinical examination confirmed the radiographic diagnosis of a cervical root perforation, subsequent to inflammatory external root resorption. This case provided the flux signal with the highest Mean Flux value in the L.D.F. Intermediate Vitality group, and the tracing is shown in Figure 6.1.

Fifty seven pulpectomies were carried out where the L.D.F. classification was Non-vital. In all cases, access to each pulp chamber was obtained without local anaesthesia and without the patient reporting pain. In all cases the contents of the coronal pulp chamber were found to be non-sensitive and non-perfused. In one case the dental pulp in the radicular canal was very sensitive to touch and required infiltration local anaesthesia for extirpation, although the pulp occupied only a quarter of the cross sectional area of the root canal, was grey in colour and was reported as partially necrotic following histological examination. However, in one case there was

Table 6.5 Laser Doppler Flowmetry classification of pulpal status, and clinical findings on pulpectomy of 67 permanent anterior teeth.

	Clinical findings on pulpectomy.			
L.D.F. classification	Vital pulp	Autolysed pulp	Necrotic pulp	Necrotic coronal, perfused apical pulp
L.D.F. Intermediate Vitality (n=10)	0	2 (20%)	5 (50%)	3 (30%)
L.D.F. Non-vital (n=57)	0	23 (40%)	30 (53%)	4 (7%)

profuse haemorrhage from the radicular canal and the pulpectomy was continued following infiltration local anaesthesia.

Further analysis of the data in Table 6.5 was carried out to determine whether necrotic dental pulps with some perfusion in the apical part of the root canal were more likely to be classified as L.D.F. Intermediate Vitality rather than L.D.F. Non-vital. The sample of autolysed dental pulps were combined with the necrotic dental pulps to form a group for which there was no clinical evidence of perfusion. This was compared with the group with perfused apical pulps, using Fischer's Exact test. It was found that there was a significantly increased chance of finding perfusion in the apical part of a necrotic pulp with an L.D.F. Intermediate Vitality classification as compared with L.D.F. Non-vital ($p<0.008$).

6.4.2 Discussion

The study found that at the macroscopic level, pathological change may not affect the pulp uniformly. Although all 67 teeth with an L.D.F. classification of Intermediate Vitality or Non-vital were found clinically to have a completely necrotic coronal pulp, 10% (n=7) were found to have some perfusion in the radicular canal. As might be expected, there was a significantly increased chance of these teeth being classified L.D.F. Intermediate Vitality due, probably, to some apical blood flow being included within the measuring volume of the coronally placed laser doppler flowmetry probe. However, four of the teeth with some perfusion in the apical half of the root canal gave flux signals which fulfilled the requirements for being classified L.D.F. Non-vital. It would appear, therefore, that the flux signal obtained with laser doppler flowmetry is principally derived from the coronal portion of the dental pulp. Revascularisation usually proceeds from the apex of a tooth (Section 2.3.6), and laser doppler flowmetry may, therefore, only indicate that a revascularising dental pulp is vital when the process is nearly complete.

6.5 HISTOPATHOLOGY OF DENTAL PULPS EXTIRPATED FOLLOWING LASER DOPPLER FLOWMETRY

6.5.1 Results

Sixty seven permanent anterior teeth were subject to pulpectomy in this study. In 25 teeth (37% of the sample) the pulpal tissue had undergone complete autolysis and there was, therefore, no tissue available for histological examination. For the remaining 42 teeth, sufficient pulpal tissue for histological examination was retrieved from 31 teeth. All tissue samples were reported on histological examination as being either partially or totally necrotic. The distribution of partial and total pulpal necrosis between the L.D.F. classification groupings of Intermediate Vitality and Non-vital is shown in Table 6.6. Further analysis of the data, using Fischer's Exact test, found no significant difference in the distribution of partial and total pulpal necrosis between the L.D.F. classifications of Intermediate Vitality and Non-vital. The histopathology was generally non-specific, and signs of ischaemic infarction (see Section 6.4.2) were reported in only four specimens.

6.5.2 Discussion

All dental pulps with a classification of L.D.F. Intermediate Vitality or L.D.F. Non-vital were found on histology to be either partially or totally necrotic. This indicated that the L.D.F. classification criteria used to classify the flux signal allowed the identification of dental pulps which were at least partially necrotic with a specificity of 1.0. The study found that the histopathology of pulpal necrosis following traumatic injury was non-specific and was not always homogenous throughout a specimen, a finding supported by other studies (Chirnside, 1957; Arwill, Henschen & Sundwall-Hagland, 1967; Cipriano & Walton, 1986 and Andreasen, 1988). This may due to the variety of mechanisms by which dental trauma can cause pulpal disease (Section 2.3).

Table 6.6 Histopathology findings on extirpated dental pulps with Laser Doppler Flowmetry classification

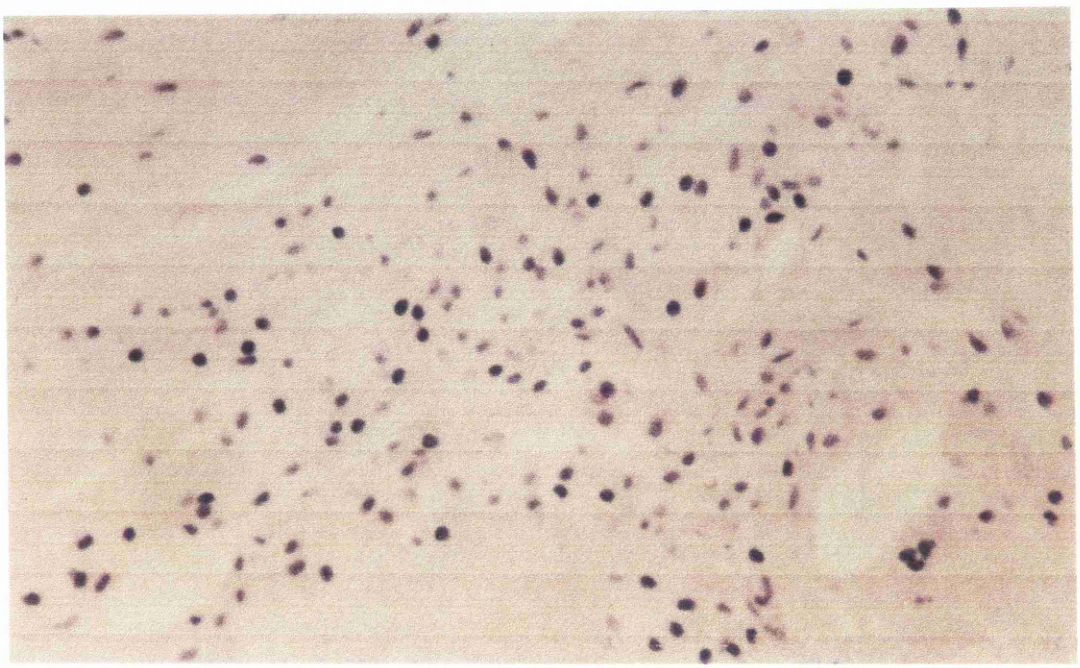
	Histopathology diagnosis		
L.D.F. classification	Vital	Partial necrosis (%)	Total necrosis (%)
L.D.F. Intermediate Vitality (n=5)	0	2 (40%)	3 (60%)
L.D.F. Non-vital (n=26)	0	8 (31%)	18 (69%)

Stanley *et al.* (1978) reported the classic signs of ischaemic infarction (extravasation of erythrocytes, nuclear degeneration, 'ghosting' of blood vessels and eosinophilic staining of the entire specimen) in a sample of 150 dental pulps from traumatised incisors. However, Cipriano *et al.* (1986) looked for ischaemic infarction as described by Stanley *et al.* (1978) in a prospective study on the dental pulps from 42 traumatised incisors and reported it in only 12% of the sample. In the present study it was only reported in 13% of the sample.

The mechanism by which some pulpal cells can apparently survive in the absence of pulpal perfusion is unclear. Some studies have reported that where there is patchy cell survival, it is most pronounced in the apical portion of the dental pulp (Ohman, 1965; Hasselgren *et al.*, 1977 and Andreasen, 1988). However, in a sample of five partially necrotic pulps from the present study where the orientation of the pulp could be reliably identified, the necrosis was found to be less extensive in the coronal section of the pulp in four of the samples. An example is shown in Figure 6.6. This surprising finding may be due to lateral root canals allowing diffusion of nutrients down concentration gradients. Lateral canals frequently occur in anterior teeth; De Deus (1975) reported lateral canals in 19% of a sample of 37 permanent maxillary central incisors, Hession (1977) found them in 43% of a sample of 14 of the same teeth and Goldberg *et al.* (1986) reported lateral canals in 64% of a sample of 22 permanent anterior teeth. However, the rate of diffusion through a canal varies with the fourth power of its radius. The apical foramen is invariably wider than lateral canals and would, therefore, be expected to allow more diffusion, and greater cell survival, in the apical pulp.

Pulpal tissue has a low level of acid hydrolase activity which can delay the signs of necrosis (Hasselgren, Larsson & Lundquist, 1977). In addition, it is possible that infarcted cells in the coronal portion of the pulp are relatively isolated from access by inflammatory cells from the systemic circulation. It is, therefore, possible that pulpal

a) Coronal portion of dental pulp



b) Apical portion of dental pulp

Figure 6.6 Photomicrographs of the dental pulp of a traumatised maxillary central incisor: (a) coronal pulp, (b) apical pulp. (Haematoxylin and eosin stain, original magnification 160X).

cells which appear vital histologically are moribund. Hasselgren *et al.* (1977) demonstrated an absence of cellular respiratory enzyme activity in infarcted pulp tissue which histologically appeared vital. Whether the cells might be capable of normal function on resumption of perfusion is not known at present. It is, therefore, difficult to define histological criteria for classifying a dental pulp as vital. The only certainty is that a necrotic pulp is non-vital.

Pulpal necrosis may be reversible (Section 2.3.6), although the criteria for assessing whether the necrosis is reversible or not have yet to be agreed. Andreasen (1988) defined the presence of pulpal infection as indicating irreversible pulpal disease, although the potential for pulpal tissue to overcome infection was discussed in Section 2.3.5. Dummer *et al.* (1980) defined the presence of necrosis in any part of a dental pulp as indicating irreversible pulpal disease. However, the study on reimplanted human premolars by Ohman (1965) demonstrated that necrotic pulpal tissue could be replaced by vital tissue growing in from the periodontal ligament. Therefore, even histological confirmation of a clinical diagnosis of pulpal necrosis does not necessarily indicate that a pulpectomy was justified.

6.6 COMPARISON OF LASER DOPPLER FLOWMETRY ASSESSMENT OF PULPAL STATUS WITH OTHER DIAGNOSTIC METHODS.

6.6.1 Results

The results of current pulpal diagnostic tests when applied to the vital and non-vital teeth test groups are shown in Table 6.7. Some current pulpal diagnostic tests require a subjective response from the patient (notably sensibility testing and percussion). Therefore, to avoid bias in the results, only one non-vital tooth per patient was included in the non-vital group. This reduced the sample size of the non-vital group from 67 teeth to 55 teeth. The comparison group of 84 vital teeth was already based on one tooth per patient. The results are reported as test sensitivity and

Table 6.7 Standard pulpal diagnostic tests for L.D.F. Non-Vital (L.D.F. Non-vital + L.D.F. Intermediate Vitality) and L.D.F. Vital teeth (n=sample size).

Diagnostic test	L.D.F. classification Non-vital (sensitivity)	L.D.F. classification Vital (specificity)
History of pain	0.16 (n=55)	0.99 (n=84)
Alveolar tenderness	0.23 (n=43)	1.0 (n=43)
Alveolar sinus	0.13 (n=55)	1.0 (n=83)
Tenderness to percussion	0.35 (n=54)	0.99 (n=76)
Crown colour	0.34 (n=53)	1.0 (n=79)
Trans illumination	0.49 (n=51)	1.0 (n=77)
Mobility	0.10 (n=41)	1.0 (n=50)
Ethyl chloride	0.92 (n=53)	0.89 (n=81)
Electric pulp test	0.87 (n=53)	0.96 (n=83)
Periapical radiolucency	0.36 (n=47)	0.97 (n=64)
Root apex resorption	0.14 (n=53)	0.98 (n=66)
External root resorption	0.13 (n=53)	1.0 (n=66)

test specificity, with the method of calculation having been described in Section 6.5.2. The test sensitivities are derived from the 55 non-vital teeth and the test specificities from the 84 vital teeth. Column totals may vary between rows either because the criteria for applying that test to the tooth could not be met (Section 6.1) or because, unfortunately, the results of that test were not recorded.

The specificities of the diagnostic tests involving history and examination, and radiographic examination, were very high, being 0.97 or better. The sensitivities of the same tests were much lower; the least sensitive signs of pulpal non-vitality were increased mobility, present in 10% of non-vital teeth and an alveolar sinus, present in only 13% of cases. A history of pain, tenderness of the alveolus to palpation, tenderness of the tooth to percussion, radiographic signs of pulpal necrosis and coronal discolouration to reflected light were individually present in 36% or less of non-vital teeth. Transillumination was the most sensitive of this group of tests but the sensitivity of 0.49 meant that about half of the sample of 55 non-vital teeth were not demonstrating discolouration.

The specificities of tests of pulpal sensibility were slightly lower than for the other tests, being 0.96 for electric pulp testing and 0.89 for ethyl chloride. However, the sensitivities of these tests showed a marked increase compared with the other methods of diagnosing pulp vitality, with electric pulp testing having a sensitivity of 0.87 and ethyl chloride a sensitivity of 0.92. Of the three vital teeth which gave a negative response to the electric pulp test, radiographs were available for two and both had apical root foramina $\geq$ 1.8 mm. Of the nine vital teeth which did not respond to testing with ethyl chloride, radiographs were available for eight and of these four had apical root foramina $\geq$ 1.8 mm. There was radiographic evidence of the apical root development of 75 of the 84 vital teeth and of these, only 11 (15% of the sample) had apical root foramina $\geq$ 1.8 mm.

On the data set available, forward stepwise regression analysis was carried out to determine which combinations of standard pulpal diagnostic tests most reliably

discriminated between vital and non-vital dental pulps. Analysis of the clinical tests (excluding radiography) indicated that the most reliable combination of tests was electric pulp testing and a history of pain. This combination of tests had a specificity of 0.98. However, the sensitivity was only 0.16. Including radiography in the analysis resulted in electric pulp testing, presence of a radiographic periapical area, presence of tenderness in the alveolar sulcus and a history of pain being selected as the most reliable tests, with a specificity of 0.98 and a sensitivity increased to 0.69. However, this mathematical combining of various diagnostic techniques did not increase the sensitivity of the tests to that of electric pulp testing alone but it did indicate that radiography should be considered as a valuable pulpal diagnostic aid.

6.6.2 Discussion

The sensitivities of the pulpal diagnostic tests involving history, clinical examination and radiographic examination were low, making them unreliable for the detection of pulpal necrosis. However, the very high specificities of these tests meant that they would be very unlikely to incorrectly diagnose a vital dental pulp as being necrotic. With the sensitivity and specificity of diagnostic tests often inversely related, this balance of high specificities against lower sensitivities would usually be to the patient's advantage (Section 2.4.2). However, there will be occasions where it will be essential to have a reliable diagnosis, such as with the medically compromised patient or the patient with learning difficulties. In addition, effective treatment planning is dependent on reliable diagnostic information. The low sensitivities of these tests was, therefore, disappointing.

The most reliable diagnostic tests were those testing pulpal sensibility, where a small fall in specificity compared with other tests was balanced by a much larger rise in sensitivity. With regard to the fall in specificity of these tests, it was of interest to note the association between a lack of patient response to sensibility testing of vital teeth and immature root formation. This finding is in agreement with other studies (Section 2.7.3).

The sample of teeth subject to pulpectomy were diagnosed as having irreversible pulpal necrosis based on the results of at least two standard diagnostic tests. It is likely, therefore, that the sensitivities of the standard diagnostic tests reported in this sample are higher than in a random sample of traumatised teeth, due to pulpectomy being postponed until there was more than one sign of pulpal necrosis. This was, however, in line with current guidelines (Andreasen, 1988).

6.7 CONCLUSIONS

Flux signals obtained by laser doppler flowmetry of 67 non-vital dental pulps and 84 vital dental pulps were classified using the following L.D.F. classification criteria:

- L.D.F. Vital - Mean Flux ≥ 7.0 P.U. and Amplitude of Slow Wave Vasomotion ≥ 1.6 P.U..
- L.D.F. Intermediate Vitality - Mean Flux ≥ 7.0 P.U. and Amplitude of Slow Wave Vasomotion < 1.6 P.U..
- L.D.F Non-vital - Mean Flux < 7.0 P.U..

All 67 dental pulps classified as L.D.F. Non-vital or L.D.F. Intermediate Vitality were found on pulpectomy to have insensitive, necrotic coronal pulps. Seven pulps (10% of the sample) were found clinically to have some perfusion in the apical part of the canal, and these pulps were significantly more likely to be classified as L.D.F. Intermediate Vitality rather than L.D.F. Non-vital. All the flux signals from the 84 vital dental pulps satisfied the criteria for classification as L.D.F. Vital. For this sample of teeth, therefore, the L.D.F. classification criteria demonstrated a sensitivity and a specificity of 1.0, making laser doppler flowmetry a more reliable pulpal diagnostic technique than any of the other pulpal diagnostic methods in current use.

The finding that sensibility testing was the most reliable of the standard pulpal diagnostic tests, with a sensitivity and specificity approaching that of laser doppler flowmetry, is of interest but it must be viewed in the context of the sample on which

the finding was based. Although the pulpal necrosis of the non-vital teeth in the sample was caused by dental trauma, the sample should not be regarded as representative of traumatised teeth in general. Diagnostic data were only collected from the non-vital teeth when irreversible pulpal necrosis was indicated by at least two standard diagnostic tests (Andreasen, 1988). It would be expected, therefore, that diagnosis of pulpal necrosis might be delayed until these clinical signs and symptoms had developed. Therefore, the standard diagnostic tests may show decreased sensitivity when applied to non-vital dental pulps where the development of pulpal necrosis had only recently occurred. In addition, although the standard diagnostic tests showed high specificities, the tests were only applied to untraumatised vital teeth. It is possible that there may be a reduction in specificities if the tests were applied to a sample of traumatised vital teeth (Section 2.7.4). A longitudinal study of traumatised permanent anterior teeth which investigates these points will be reported in Chapter 7.

CHAPTER 7

A LONGITUDINAL STUDY OF THE VITALITY OF A SAMPLE OF TRAUMATISED ANTERIOR TEETH AS ASSESSED USING LASER DOPPLER FLOWMETRY.

7.1 INTRODUCTION AND AIMS

Pulpal necrosis is a well known complication of traumatic injury of teeth. What is still unclear, however, is the temporal relationship between traumatic injury and the development of pulpal necrosis. There is, therefore, uncertainty as to the time interval following traumatic injury before which pulpal vitality can be regarded as established. Suggested intervals vary between three months to over two years (Section 2.3.3). This uncertainty is largely due to the unreliability of current methods of assessing dental pulp vitality. It is possible that the apparent late development of pulpal necrosis in traumatised teeth is, in fact, due to delay in the diagnosis of a pulpal necrosis which actually developed at the time of trauma. The new technique of laser doppler flowmetry is a reliable method of determining dental pulp vitality of anterior teeth (Chapter 6). Although time consuming for both patient and clinician, the technique, uniquely for a pulpal diagnostic test, allows an objective, non-invasive, direct assessment to be made of the vitality of a dental pulp, without requiring a subjective response from the patient or a subjective assessment by the clinician. Its use in a longitudinal study of traumatised teeth would allow an assessment to be made of the timing of the development of pulpal necrosis relative to the time of injury. In addition, the relative reliability of standard methods of assessing dental pulp vitality could also be determined.

Current methods of assessing dental pulp vitality can be unreliable. The study reported in Chapter 6 found that while laser doppler flowmetry (L.D.F.) had a diagnostic sensitivity and specificity of 1.0 when assessing dental pulp vitality, the standard tests were not as dependable. Tests of pulpal sensibility were the most

reliable of these tests, with sensitivities of 0.87 for the electric pulp test and 0.92 for ethyl chloride, and with specificities of 0.96 for the electric pulp test and 0.89 for ethyl chloride. The remaining standard tests showed high specificities (range 0.96-1.0) but low sensitivities (range 0.10-0.49). However, the sample of teeth from which these data were obtained were selected in order to validate the L.D.F. classification criteria and were not truly representative of a sample of traumatised teeth. This sample, termed the Comparison Group, was described in Section 6.2.

The non-vital teeth (used to determine test sensitivity), although devitalised as a consequence of trauma, were only included in the sample when at least two standard diagnostic tests indicated pulpal necrosis and pulpectomy was planned. This might have artificially increased the sensitivity of the tests, due to delaying the diagnosis of pulpal necrosis and allowing more time for bacterial infection to occur, radiographic signs to develop or discolouration to appear.

The sample of vital teeth (used to determine test specificity) had never been subjected to traumatic injury. There are indications from the literature (Section 2.7.4) that standard pulpal diagnostic tests show a decrease in reliability when applied to traumatised teeth, principally with a fall in the specificity of tests of pulpal sensibility. In other words, vital traumatised teeth may fail to respond to testing with the electric pulp tester or with ethyl chloride. Although this has long been suspected (Section 2.7.4), laser doppler flowmetry is the only pulpal diagnostic test, with the exception of pulpectomy, which would allow contemporaneous confirmation of this phenomenon.

It is possible, therefore, that the test sensitivities and specificities obtained from a sample of traumatised teeth might be even lower than the values obtained from the Comparison Group. Laser doppler flowmetry, with a diagnostic sensitivity and specificity of 1.0 for dental pulp vitality, would allow a valid assessment of the reliability, that is the sensitivity and specificity, of standard pulpal diagnostic tests when assessing traumatised teeth.

In the study described in this chapter the pulpal vitality of a cohort of traumatised teeth was assessed over time, using laser doppler flowmetry as the

"benchmark" pulpal diagnostic test and standard pulpal diagnostic tests as the comparators, with the following aims:

a) to determine whether pulpal status is established at the time of trauma, or if it can change subsequent to the trauma.

b) to investigate whether the sensitivity of standard diagnostic tests fall if used to assess the vitality of a representative sample of non-vital traumatised teeth as compared with a sample composed entirely of non-vital traumatised teeth where at least two standard diagnostic tests indicate loss of vitality.

c) to investigate whether the specificity of standard diagnostic tests fall if the tests are used to assess the vitality of vital traumatised teeth as compared with vital non-traumatised teeth.

7.2 METHODS AND MATERIALS

The subjects were 44 patients attending Glasgow Dental Hospital having sustained injury to their permanent anterior teeth. Seventy nine permanent anterior teeth (from 44 patients, 23 males, 21 females, mean age 11.5 years (range 6.5-24.5)) were included in the study. Following a full history and examination, dento-alveolar injuries were classified as concussion, subluxation, lateral luxation, extrusive luxation and avulsion and dental injuries as uncomplicated crown fracture, complicated crown fracture and root fracture (W.H.O., 1966). Patient data were only included in specified trauma categories when there was no history of previous trauma and where there was documented and radiographic evidence of the type of dental injury sustained. Treatment of the acute phase of dental trauma was in line with current guidelines (Andreasen, 1981b). Patients were reviewed as indicated by their treatment needs, usually at 1, 3, 12, 24 and 52 weeks following injury. The vitality of the traumatised teeth was assessed as described in Section 6.2. The laser doppler flowmetry classification groupings of L.D.F. Intermediate Vitality and L.D.F. Non-

vital were combined to form the L.D.F. Non-vital group (Section 6.4.2). Teeth were reviewed until the pulpal diagnosis was confirmed; for a vital pulp this was by continued root growth noted on consecutive radiographs and for non-vital teeth confirmation was by pulpectomy. Pulpectomy of non-vital teeth was carried out following a diagnosis of irreversible pulpal necrosis.

The criteria for classifying a non-vital pulp as being irreversibly necrotic changed during the course of the study. At the commencement of the study, pulpal necrosis was classified as being irreversible when occurring in a tooth with a radiographically closed root apex. However, during the study it was noted that some non-vital teeth with radiographically closed apices where pulpectomy had been delayed, had gone on to revascularise. Clearly, it would have been unethical to continue with the existing criteria and they were, therefore, modified during the study to include only non-vital teeth which showed radiographic evidence of an expanding periapical radiolucency or inflammatory external root resorption.

7.3 RESULTS

Data are grouped according to the trauma categories of concussion, subluxation, luxation (extrusive and lateral luxation combined) and avulsion. The L.D.F. classifications for each tooth over the review period are shown, together with the sensitivity and specificity of standard pulpal diagnostic tests, as assessed by laser doppler flowmetry, for each trauma category. Within these categories, data are grouped into time intervals 0-4 weeks, 5-12 weeks, 13-24 weeks, 25-52 weeks and 53-72 weeks post-injury. A tooth is included in each time interval on only one occasion. For example, if a tooth had been assessed at 26 weeks and at 34 weeks following trauma, only the assessment at 26 weeks would be included in the interval 25-52 weeks. Sample sizes in some time periods may be relatively small due to the requirements of the method of calculating test specificity and sensitivity. Test specificity can only be determined from a sample of vital teeth and, therefore, following a severe injury such as luxation, relatively few vital teeth may be available for assessment. Similarly, test sensitivity can only be determined from a sample of

non-vital teeth and sample sizes for concussion injuries may be small. The relatively small sample sizes in some time intervals increase the possibility of bias in the results for that period. For example, if a sample of five teeth is used to derive a value for test sensitivity, then a single aberrant result for a tooth would affect the value obtained by 0.20. Because of this, the mean values for test sensitivity and specificity over the time period 0-72 weeks post-injury is also shown. This value, unlike the value derived for each specified time period, will contain more than one test occasion per tooth. Because of these difficulties, the results are presented using descriptive statistics only. To allow comparison, data for each standard diagnostic test obtained from the study reported in Chapter 6 is shown, and is labelled "Comparison Group".

7.3.1 Concussion injury

Nine teeth (eight patients) were included in the category of concussion injury. The L.D.F. classification for these teeth over time is shown in Figure 7.1. One tooth was classified L.D.F. Vital one week following trauma, L.D.F. Intermediate Vitality at 19 weeks and again at 40 weeks following trauma before reverting to L.D.F. Vital 60 weeks following trauma. The remaining teeth were all classified as L.D.F. Vital over the review period, which ranged between 8-80 weeks post trauma.

The data for the specificities of the standard diagnostic tests are shown in Tables 7.1-7.12. Data for the sensitivities of the tests are not shown as only one tooth was classified L.D.F. Intermediate Vitality before reverting to L.D.F. Vital, with the result that there is insufficient data from which to derive test sensitivities. The specificities of the diagnostic tests over the review period were similar to those obtained from the Comparison Group of non-traumatised vital teeth; the mean specificities over time for all the tests varying by less than 0.10 from the values obtained from the comparison group.

The false positives for pulpal necrosis indicated by the tests of pulpal sensibility, tenderness to percussion and transillumination occurred in the time intervals 0-12 weeks following trauma. All reverted to true negatives over a longer follow up period.

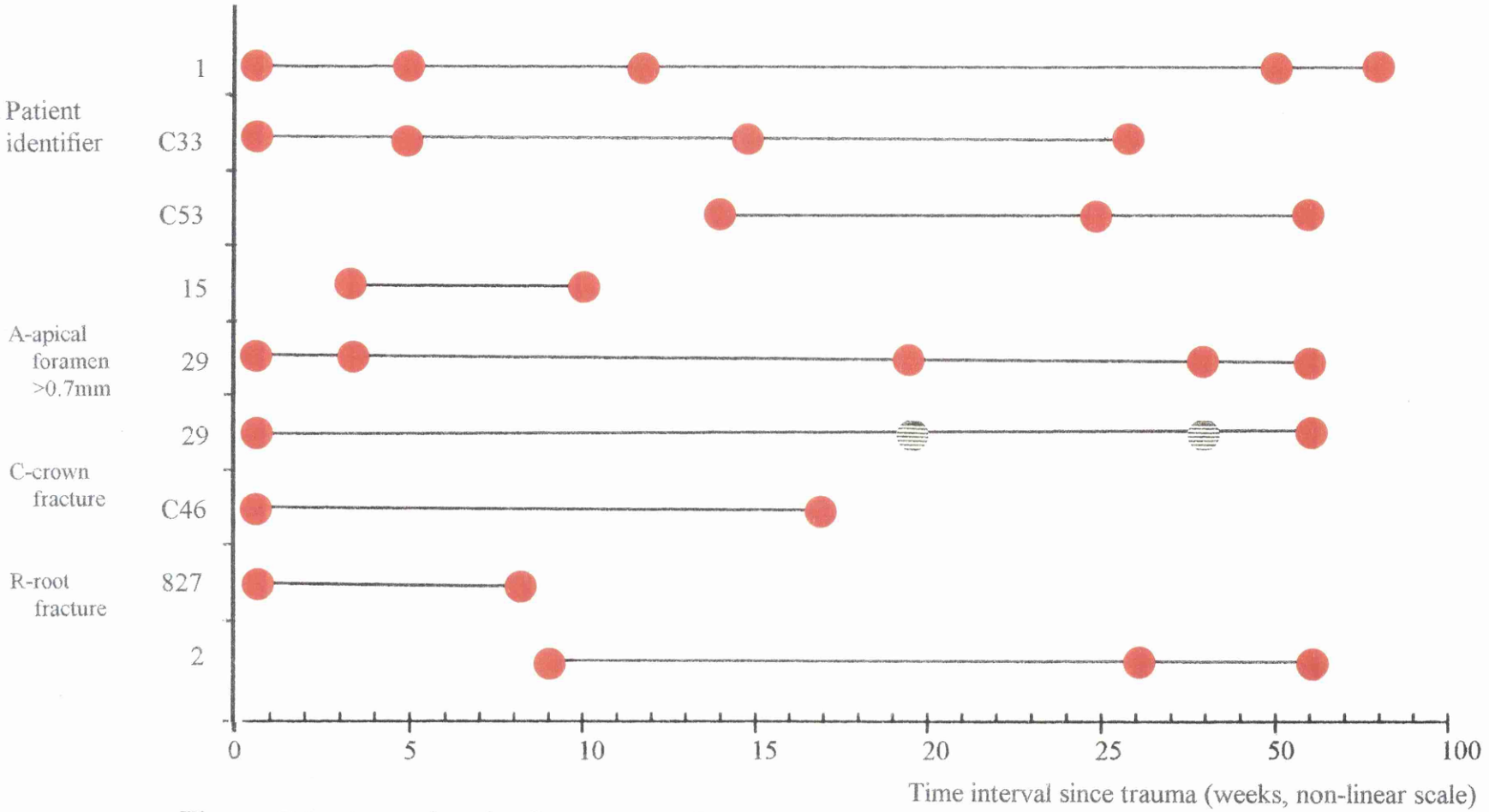


Figure 7.1 Laser doppler flowmetry (LDF) classification over time for concussed permanent incisors

Table 7.1 History of pain; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	N/A	1.00 n=4	1.00 n=4	1.00 n=4	1.00 n=5	1.00 n=17	0.99 n=84

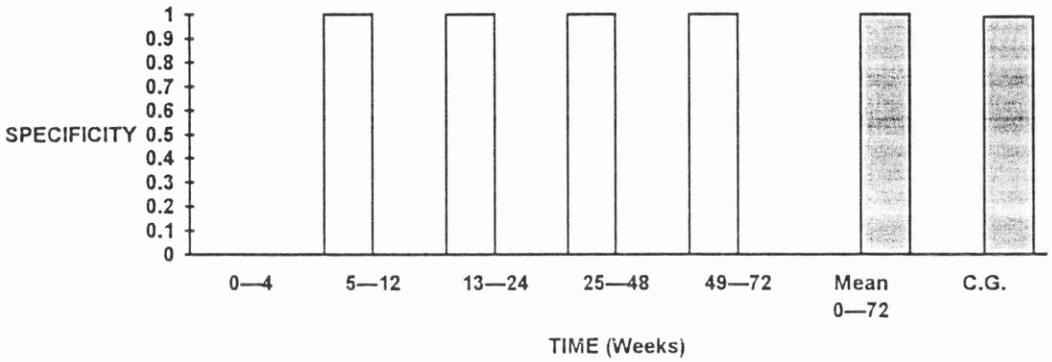


Table 7.2 Alveolar tenderness; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	N/A	1.00 n=1	1.00 n=4	1.00 n=4	1.00 n=5	1.00 n=14	1.00 n=43

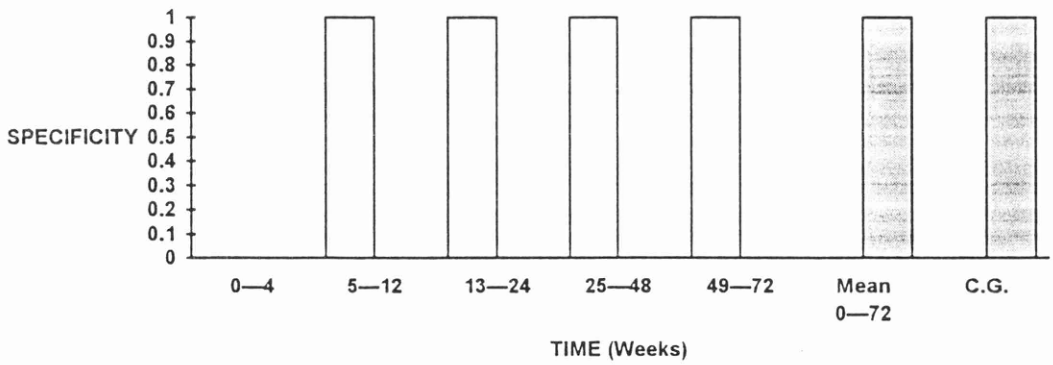


Table 7.3 Alveolar sinus; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=5	1.00 n=4	1.00 n=4	1.00 n=4	1.00 n=5	1.00 n=22	1.00 n=83

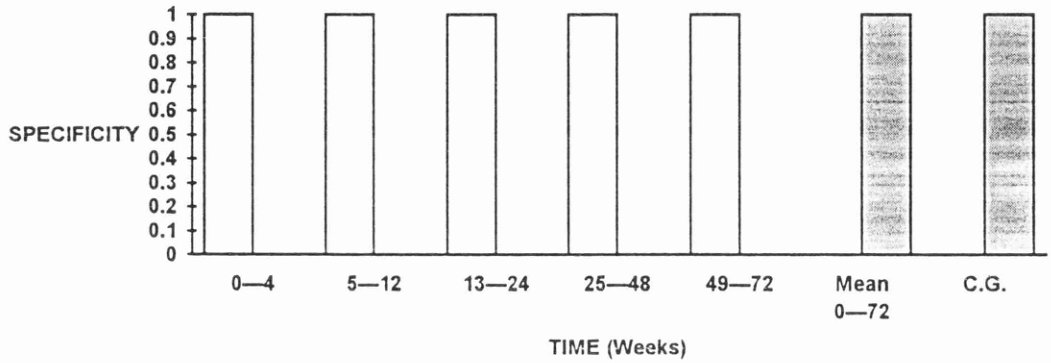


Table 7.4 Tenderness to percussion; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	N/A	0.75 n=4	1.00 n=4	1.00 n=4	1.00 n=5	0.94 n=17	0.99 n=76

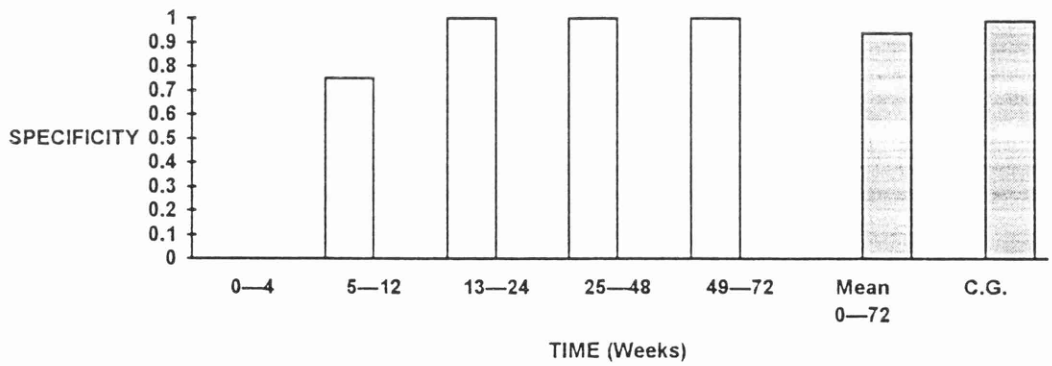


Table 7.5 Crown colour; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=5	1.00 n=4	1.00 n=4	1.00 n=3	1.00 n=5	1.00 n=21	1.00 n=79

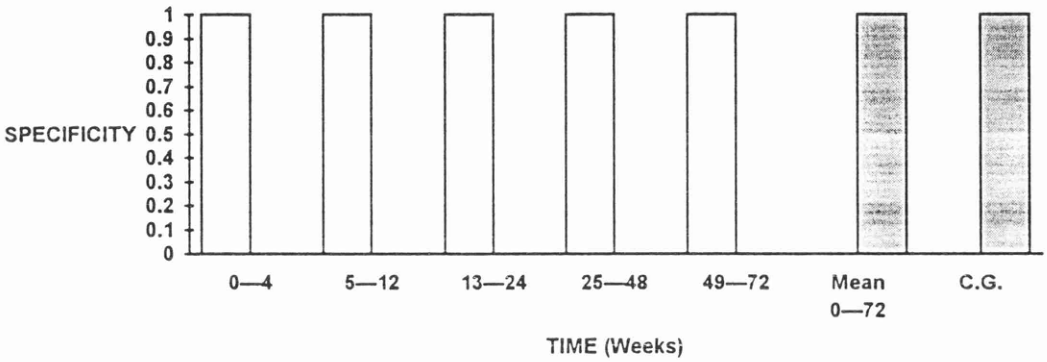


Table 7.6 Transillumination; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	0.60 n=5	1.00 n=4	1.00 n=4	1.00 n=4	1.00 n=5	0.91 n=22	1.00 n=77

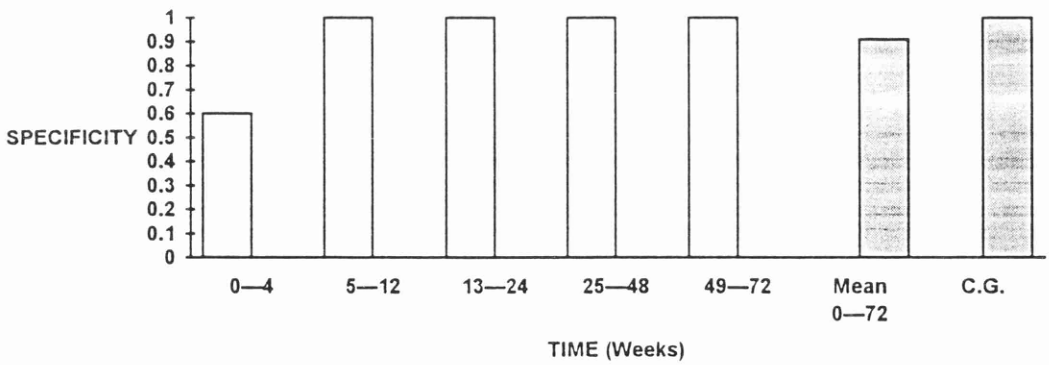


Table 7.7 Mobility; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=2	1.00 n=3	1.00 n=4	1.00 n=4	1.00 n=5	1.00 n=18	1.00 n=50

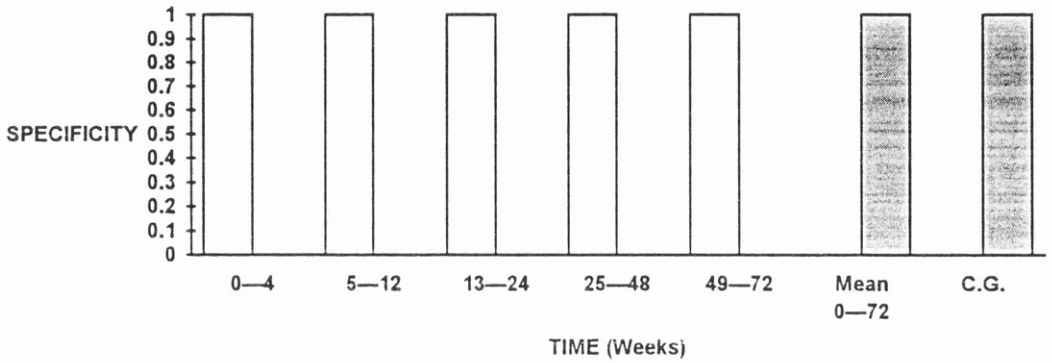


Table 7.8 Ethyl chloride; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	0.80 n=5	1.00 n=4	1.00 n=4	1.00 n=4	1.00 n=5	0.96 n=22	0.89 n=81

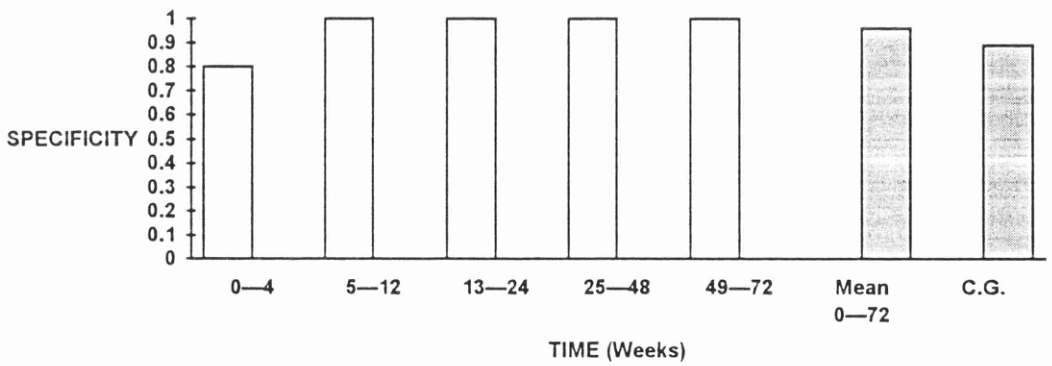


Table 7.9 Electric pulp test; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	0.60 n=5	1.00 n=4	1.00 n=4	1.00 n=4	1.00 n=5	0.91 n=22	0.96 n=83

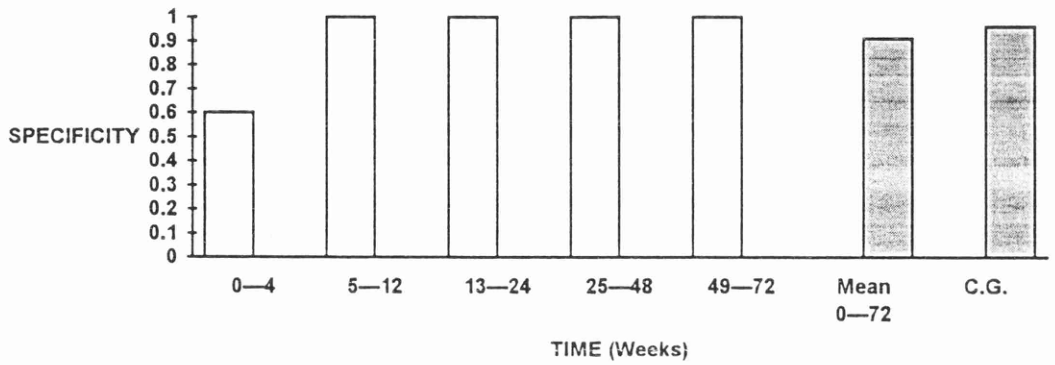


Table 7.10 Periapical radiolucency; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=4	1.00 n=3	1.00 n=1	1.00 n=3	1.00 n=5	1.00 n=16	0.97 n=64

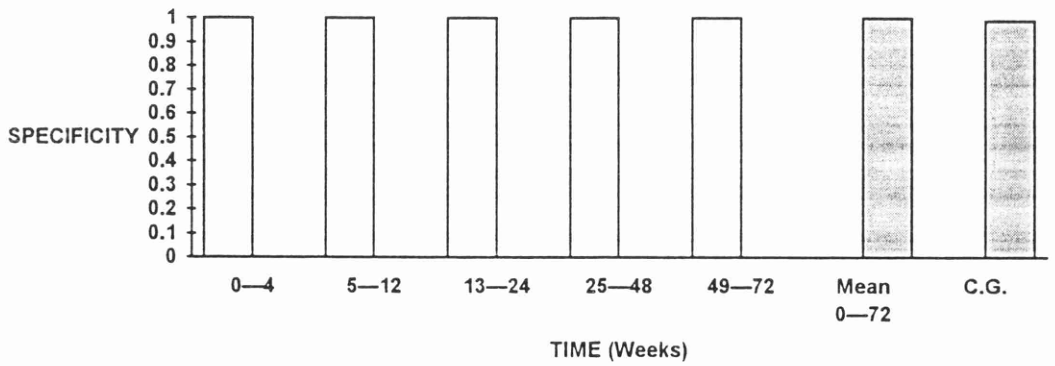


Table 7.11 Root apex resorption; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=4	1.00 n=3	1.00 n=1	1.00 n=3	1.00 n=5	1.00 n=16	0.98 n=66

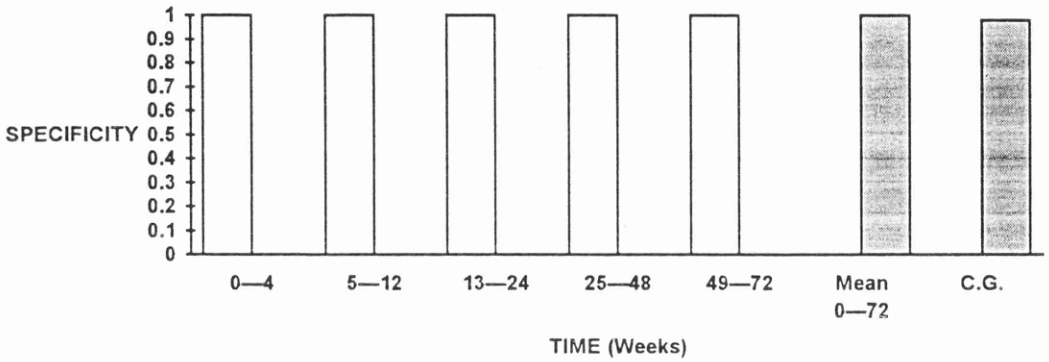
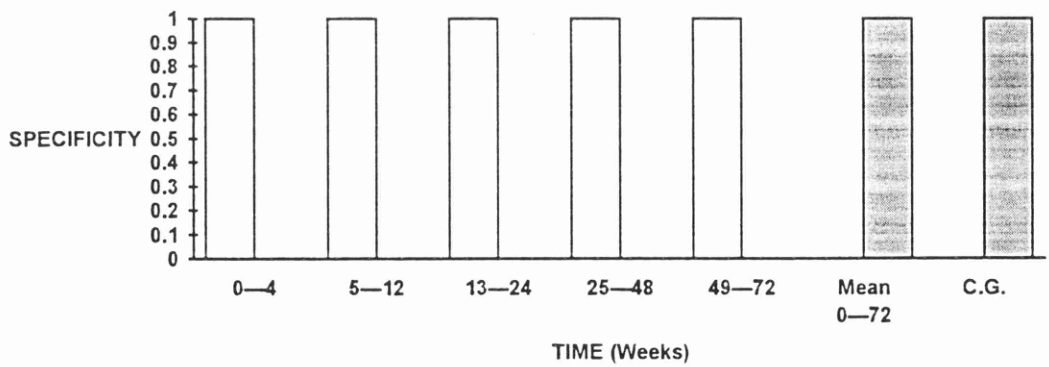


Table 7.12 External root resorption; diagnostic specificity when vitality testing concussed permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Specificity	1.00 n=4	1.00 n=3	1.00 n=1	1.00 n=3	1.00 n=5	1.00 n=16	1.00 n=66



7.3.2 Subluxation injury

Thirty one teeth (21 patients) were included in the category of subluxation injury. Eight teeth (five patients) were classified L.D.F. Vital on a single occasion but were lost to follow up. The L.D.F. classifications over time for the 23 teeth where there were two or more L.D.F. recordings are shown in Figure 7.2. Of these 23 teeth, nine teeth (39% of the sample) remained L.D.F. Vital over a review period ranging between 6-120 weeks. Five teeth (22% of the sample) remained L.D.F. Non-vital, with the diagnosis confirmed by pulpectomy for three of the teeth (the remaining two L.D.F. Non-vital teeth are still under review).

The L.D.F. classification of some teeth changed over time: of the 23 teeth for which two or more L.D.F. recordings were available, the L.D.F. classification changed for nine teeth (39% of the sample); five L.D.F. Non-vital teeth became L.D.F. Vital, while three L.D.F. Vital teeth became L.D.F. Non-vital before reverting to L.D.F. Vital. Finally, the remaining tooth was L.D.F. Non-vital, became L.D.F. Vital and then reverted to L.D.F. Non-vital and is still under review. For four of the nine teeth where the L.D.F. classification changed, the change occurred more than 12 weeks following trauma, while for two of the teeth, the change occurred more than 22 weeks following injury.

The data for the sensitivities and specificities of standard diagnostic tests are shown in Tables 7.13-7.24. The sensitivities of all standard pulpal diagnostic tests were generally either similar to or a little less than those obtained from the Comparison Group. Test sensitivities, excluding tests of pulpal sensibility and crown transillumination, were low across all the time intervals, never exceeding 0.50. Crown transillumination was a little more reliable in identifying pulpal necrosis, with a sensitivity of 0.57 during the time period 0-4 weeks and a sensitivity of 0.80 during the period 13-24 weeks. Mean sensitivities over all time periods of all tests other than those of pulpal sensibility ranged between +0.03 and -0.23 of the values obtained from the Comparison Group. Tests of pulpal sensibility showed relatively high sensitivities, generally in excess of 0.64 with the exception of a value of 0.20 (sample size five non-vital teeth) obtained for electric pulp testing during the time interval 13-24 weeks after trauma. Again, these values were generally equal to or a little lower

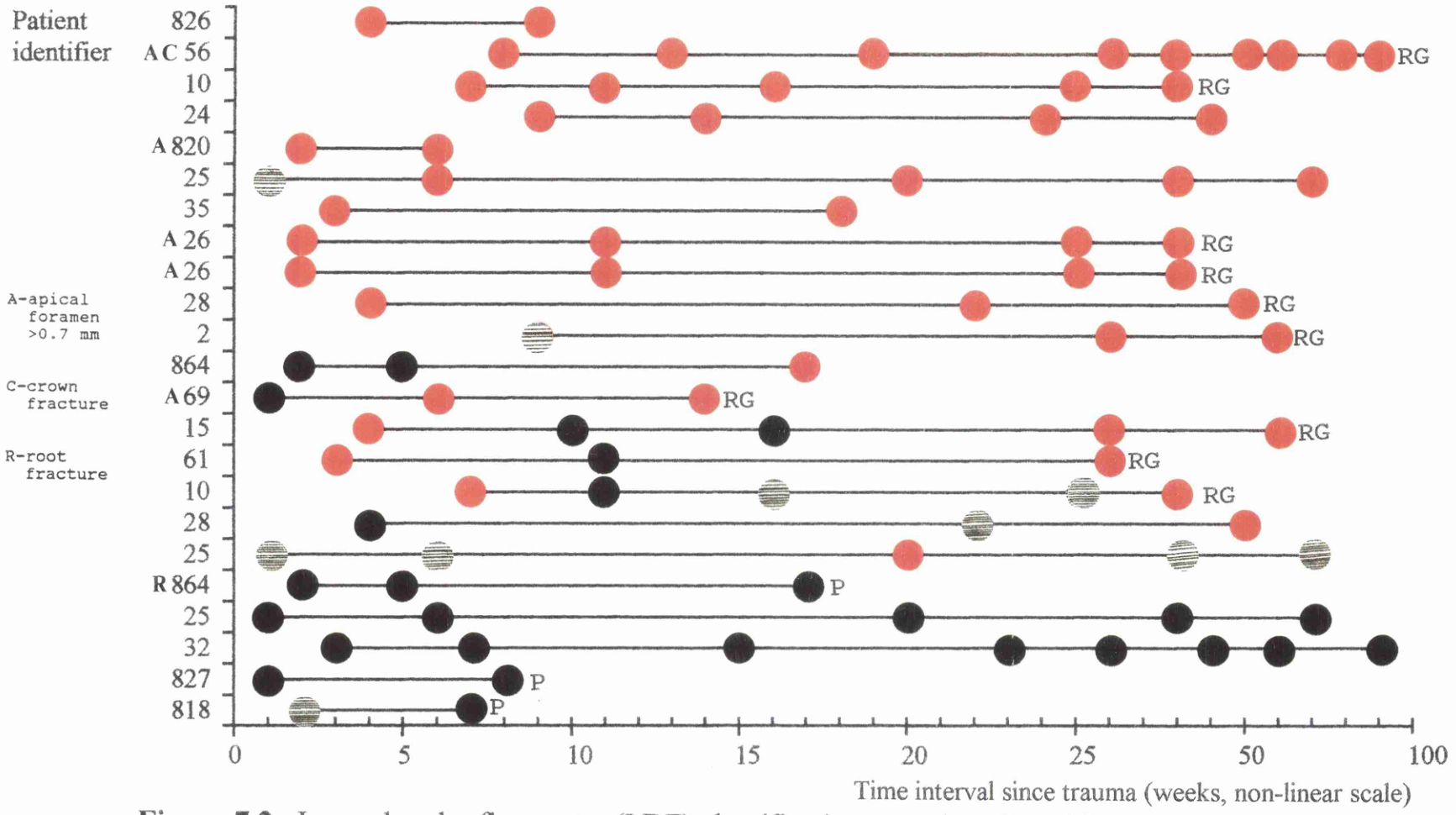


Figure 7.2 Laser doppler flowmetry (LDF) classification over time for subluxed permanent incisors

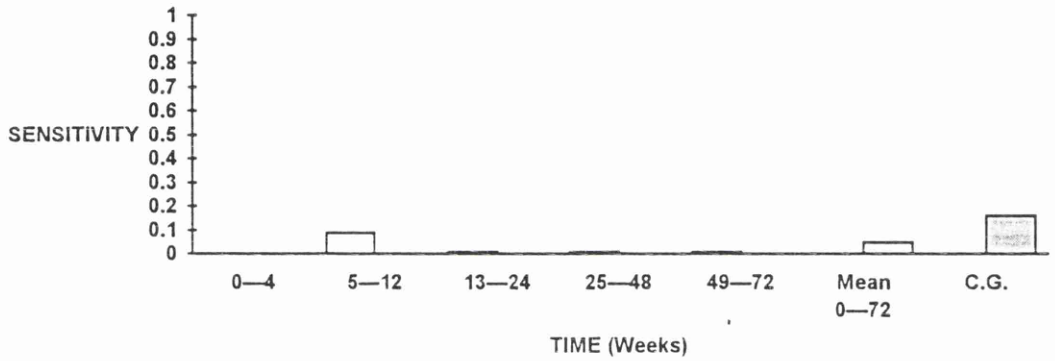
-LDF Vital, RG -classification confirmed by continued root growth

-LDF Intermediate vitality

-LDF Non-vital, P -classification confirmed by pulpectomy

Table 7.13 History of pain; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.09 n=11	0.00 n=5	0.00 n=4	0.00 n=1	0.05 n=21	0.16 n=55
Specificity	N/A	1.00 n=11	1.00 n=10	1.00 n=11	1.00 n=6	1.00 n=38	0.99 n=84



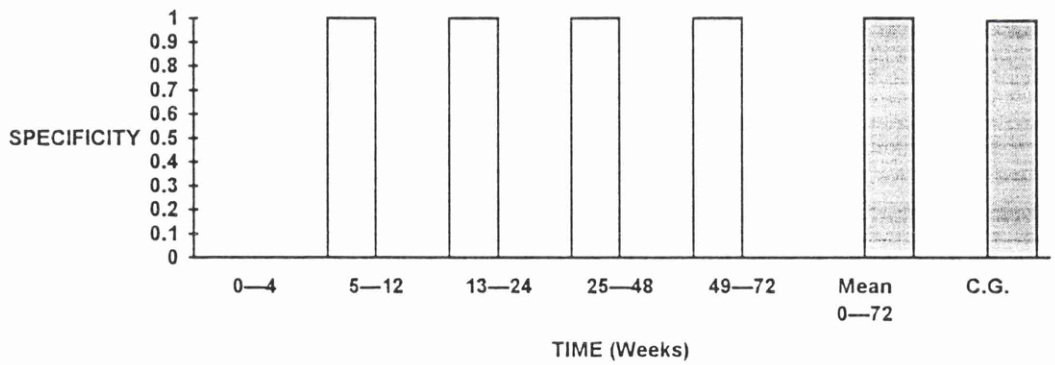
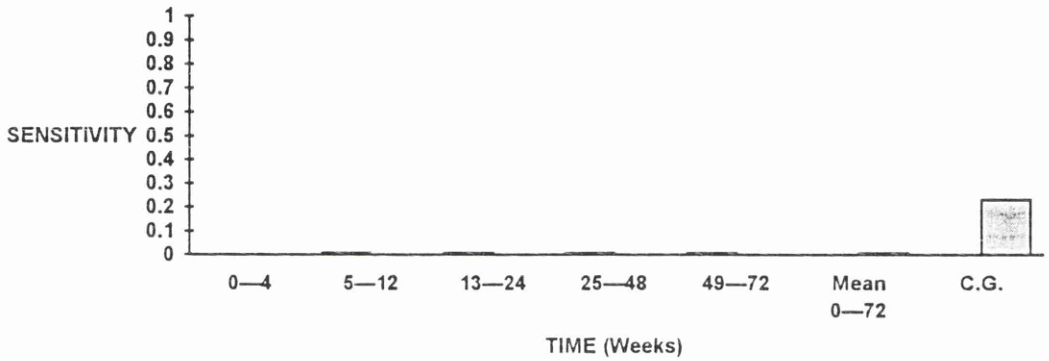


Table 7.14 Alveolar tenderness; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.00 n=3	0.00 n=2	0.00 n=2	0.00 n=1	0.00 n=8	0.23 n=43
Specificity	N/A	1.00 n=5	1.00 n=6	1.00 n=6	1.00 n=3	1.00 n=20	1.00 n=43



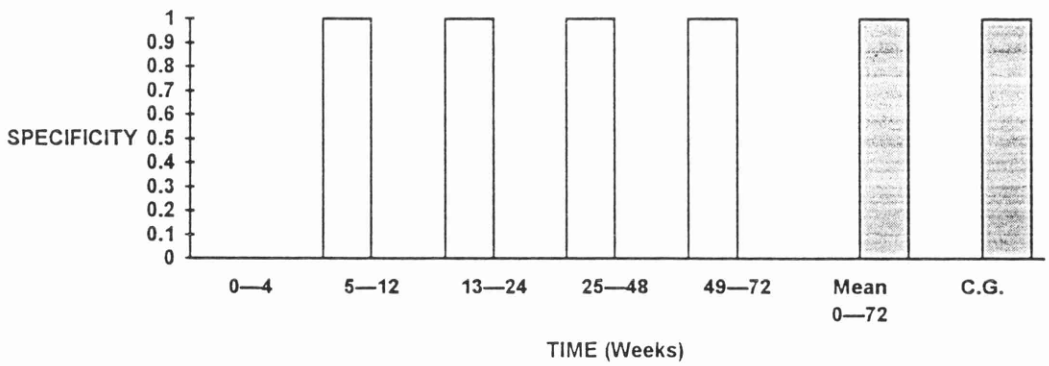
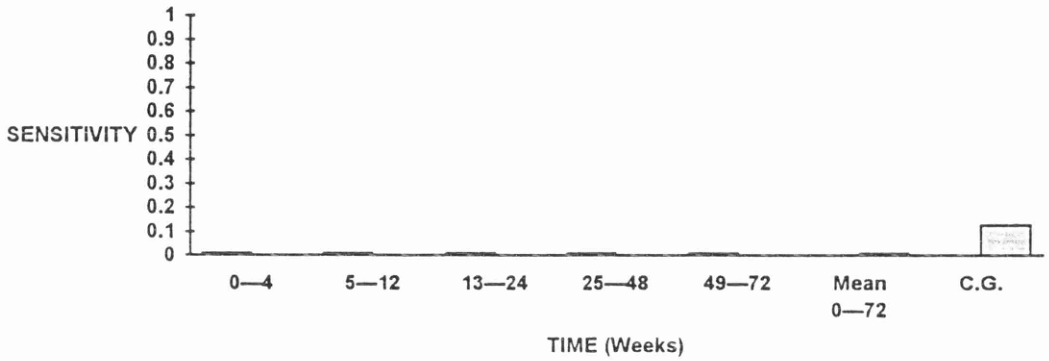


Table 7.15 Alveolar sinus; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.00 n=8	0.00 n=11	0.00 n=5	0.00 n=4	0.00 n=1	0.00 n=29	0.13 n=55
Specificity	1.00 n=14	1.00 n=11	1.00 n=10	1.00 n=11	1.00 n=6	1.00 n=52	1.00 n=83



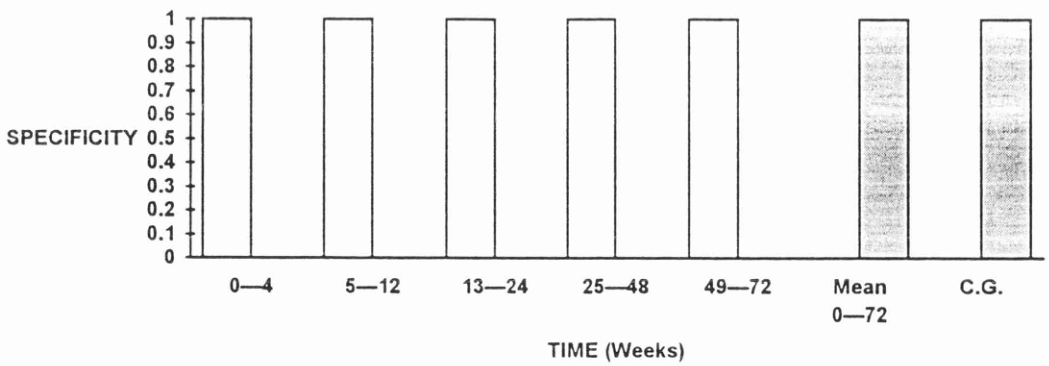
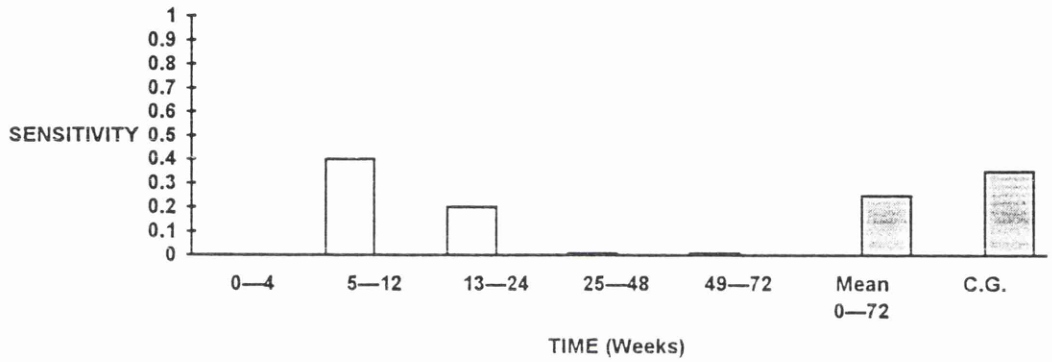


Table 7.16 Tenderness to percussion; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.40 n=10	0.20 n=5	0.00 n=4	0.00 n=1	0.25 n=20	0.35 n=54
Specificity	N/A	0.80 n=10	0.90 n=10	0.91 n=11	0.75 n=4	0.86 n=35	0.99 n=76



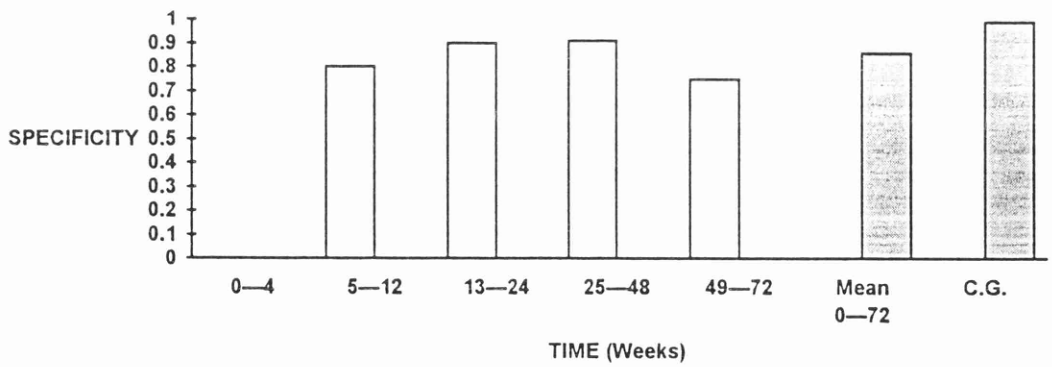
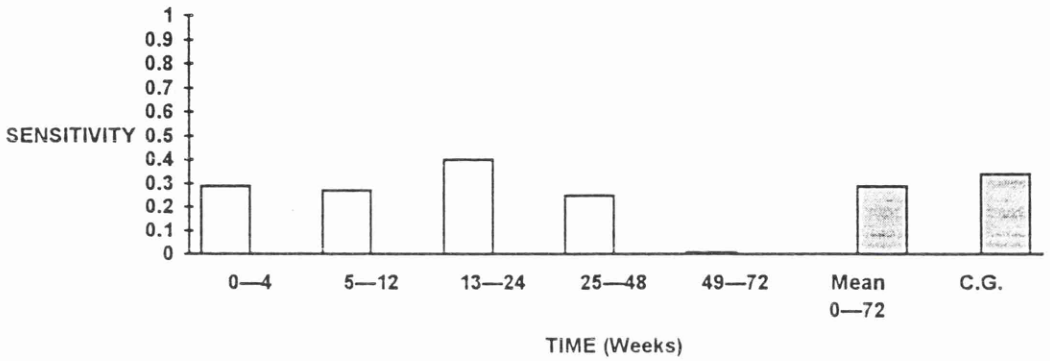


Table 7.17 Crown colour; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.29 n=7	0.27 n=11	0.40 n=5	0.25 n=4	0.00 n=1	0.29 n=28	0.34 n=53
Specificity	0.86 n=14	1.00 n=11	1.00 n=10	1.00 n=11	1.00 n=6	0.96 n=52	1.0 n=79



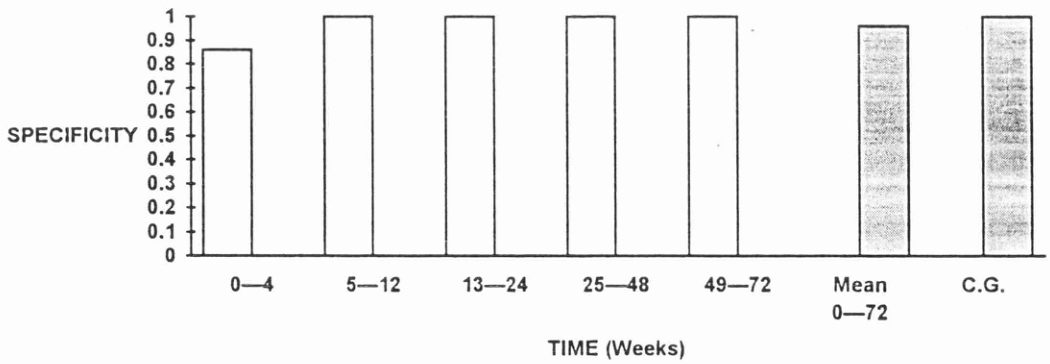
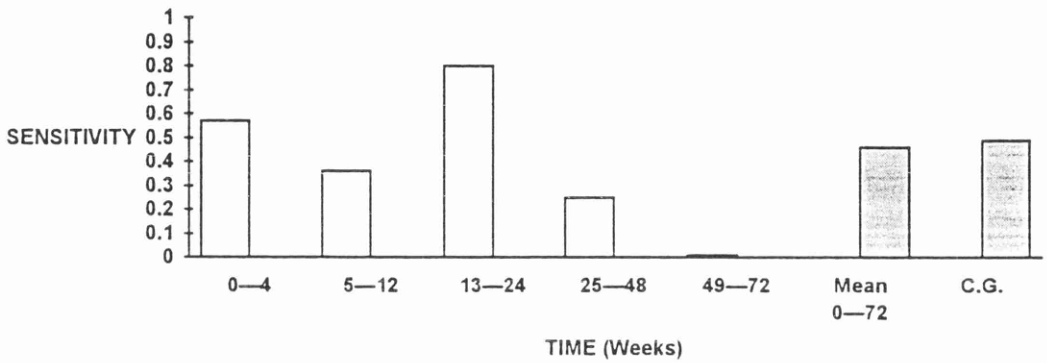


Table 7.18 Crown transillumination; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.57 n=7	0.36 n=11	0.80 n=5	0.25 n=4	0.00 n=1	0.46 n=28	0.49 n=51
Specificity	0.86 n=14	1.00 n=11	1.00 n=10	1.00 n=11	0.83 n=6	0.94 n=52	1.00 n=77



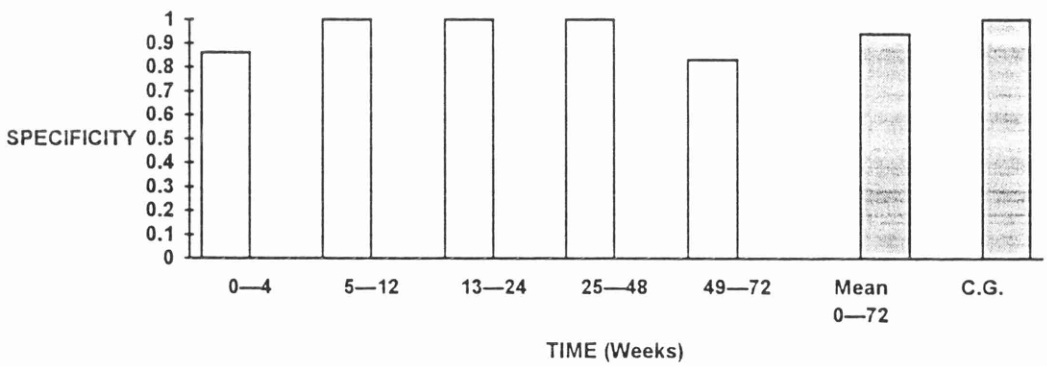
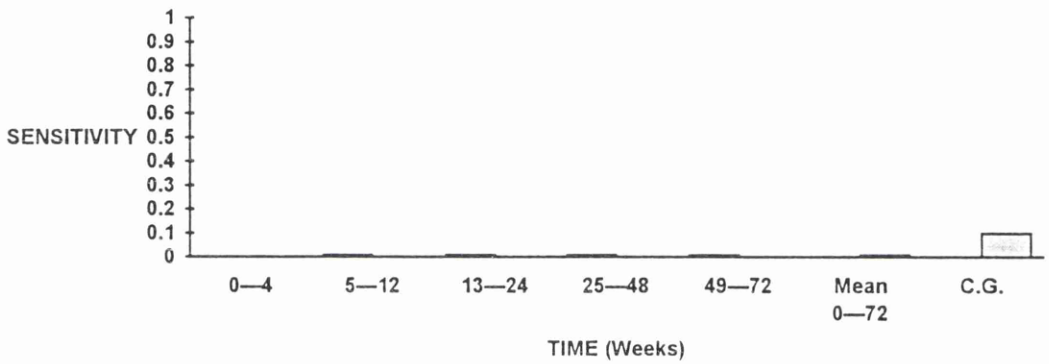


Table 7.19 Mobility; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.00 n=4	0.00 n=2	0.00 n=3	0.00 n=1	0.00 n=10	0.10 n=41
Specificity	N/A	1.00 n=7	1.00 n=5	1.00 n=6	1.00 n=3	1.00 n=21	1.00 n=50



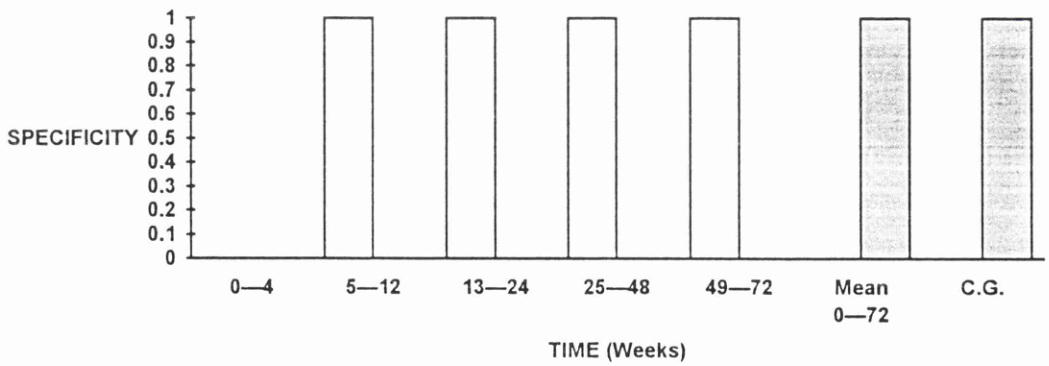
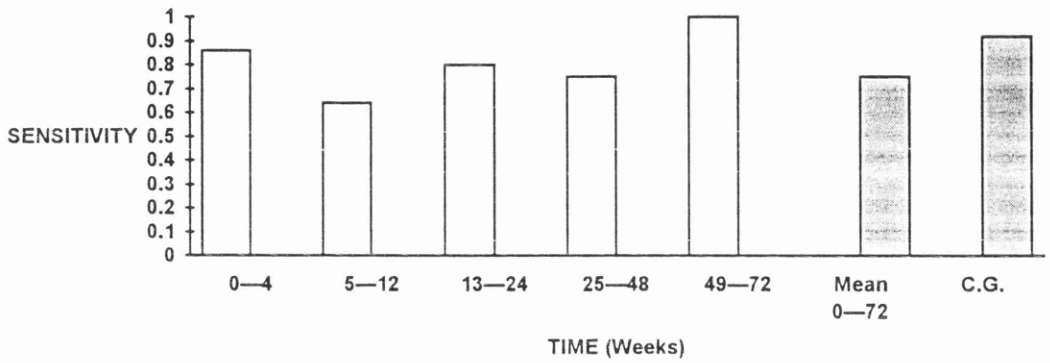


Table 7.20 Ethyl chloride; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.86 n=7	0.64 n=11	0.80 n=5	0.75 n=4	1.00 n=1	0.75 n=28	0.92 n=53
Specificity	0.79 n=14	0.50 n=10	0.44 n=9	0.18 n=11	0.67 n=6	0.52 n=50	0.89 n=81



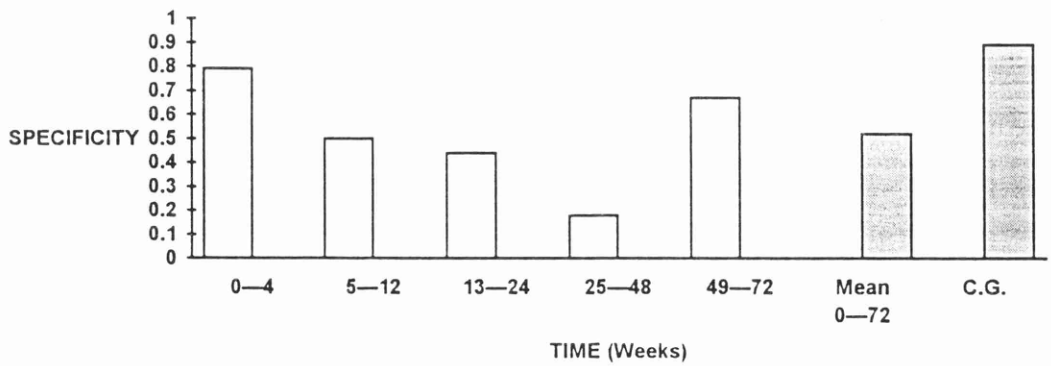
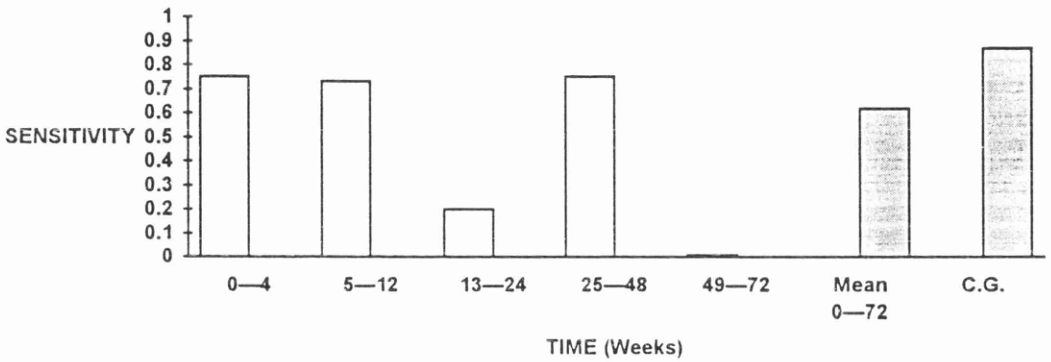


Table 7.21 Electric pulp test; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.75 n=8	0.73 n=11	0.20 n=5	0.75 n=4	0.00 n=1	0.62 n=29	0.87 n=53
Specificity	0.57 n=14	0.45 n=11	0.60 n=10	0.70 n=10	0.83 n=6	0.61 n=51	0.96 n=83



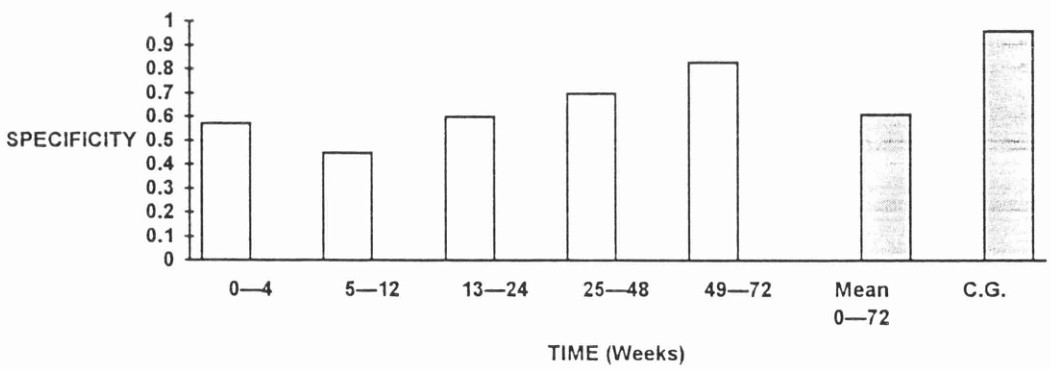
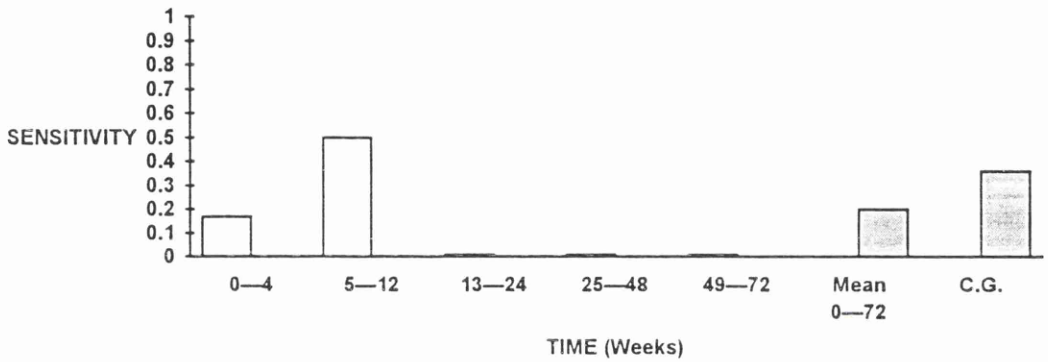


Table 7.22 Periapical radiolucency; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.17 n=6	0.50 n=6	0.00 n=4	0.00 n=3	0.00 n=1	0.20 n=20	0.36 n=47
Specificity	0.83 n=6	0.80 n=5	0.78 n=9	1.00 n=7	1.00 n=5	0.88 n=32	0.97 n=64



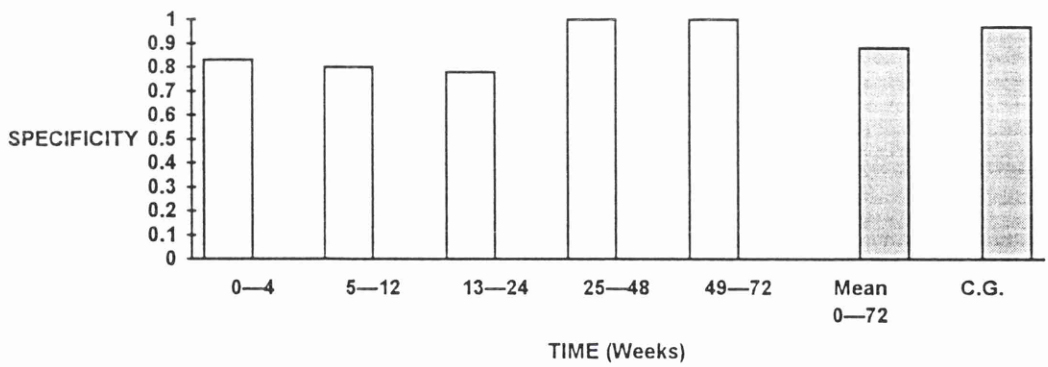
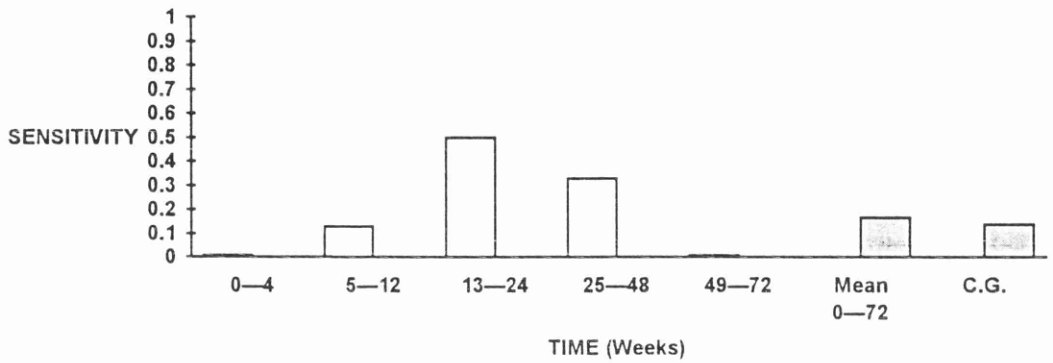


Table 7.23 Root apex resorption; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.00 n=7	0.13 n=8	0.50 n=4	0.33 n=3	0.00 n=1	0.17 n=23	0.14 n=53
Specificity	1.00 n=7	1.00 n=5	0.89 n=9	0.86 n=7	1.00 n=5	0.94 n=33	0.98 n=66



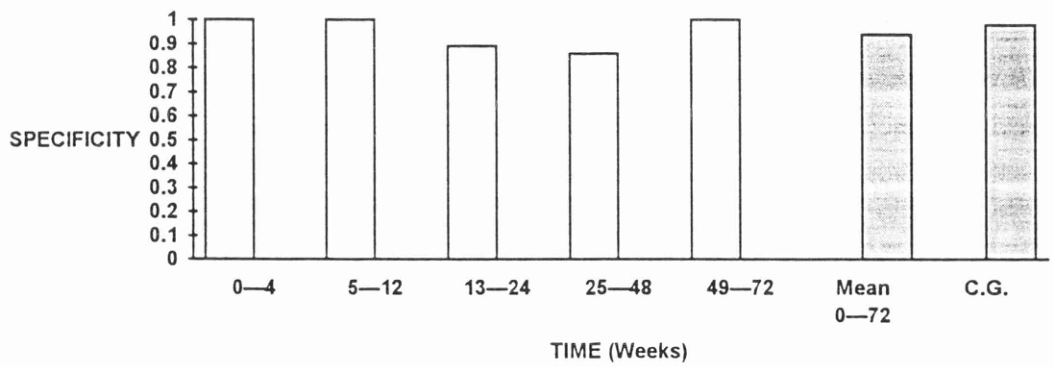
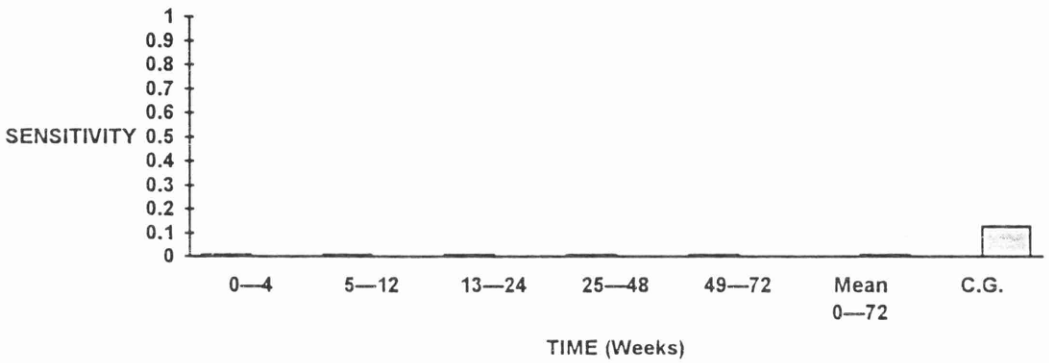
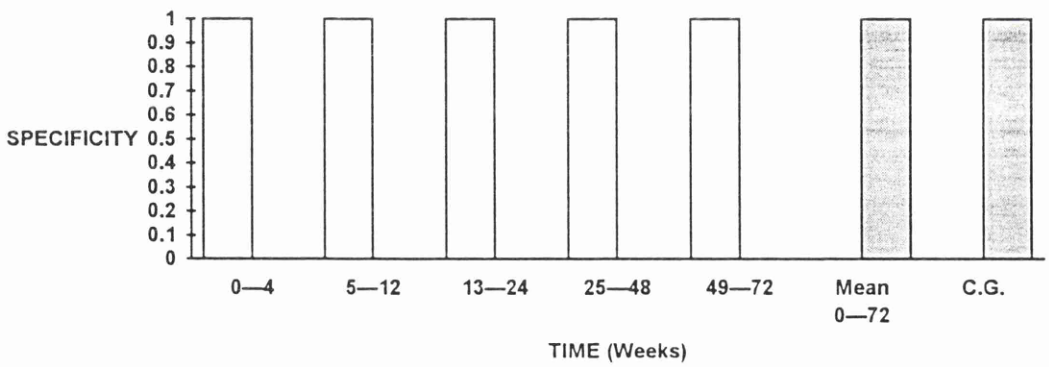


Table 7.24 External root resorption; diagnostic sensitivity and specificity when vitality testing subluxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.00 n=7	0.00 n=8	0.00 n=4	0.00 n=3	0.00 n=1	0.00 n=23	0.13 n=53
Specificity	1.00 n=7	1.00 n=5	1.00 n=9	1.00 n=7	1.00 n=5	1.00 n=33	1.00 n=66





than those obtained from the Comparison Group, with the mean values over all the time periods being less by 0.22 (ethyl chloride) and 0.25 (electric pulp testing) than the values obtained from the Comparison Group.

With the exception of tests of pulpal sensibility, the mean specificities of standard pulpal diagnostic tests generally followed the same pattern, being generally the same as or a little lower than the values obtained from the Comparison Group. Across all the time intervals, no specificity was lower than 0.24 from the value obtained from the Comparison Group, and the mean values obtained never fell by more than 0.13 from the values obtained from the Comparison Group. However, for tests of pulpal sensibility the specificities of the tests in several of the time intervals showed a comparatively greater fall when compared against the Comparison Group (see Tables 7.20 and 7.21) and the mean specificities over all the time intervals fell by 0.37 for ethyl chloride and by 0.35 for electric pulp testing.

7.3.3 Luxation injuries

Thirty one teeth (22 patients) were included in the category of luxation injury. The L.D.F. classifications over time for these teeth are shown in Figure 7.3. Five teeth classified L.D.F. Non-vital on a single occasion, within five weeks of trauma, were subject to immediate pulpectomy due to the criteria existing for pulpectomy in the early part of the study (see Section 7.2). Recordings over time were, therefore, available for 26 teeth (review period 7-110 weeks).

Of these 26 teeth, three teeth were always L.D.F. Vital while 10 of the teeth were always L.D.F. Non-vital. However, as with the sample of subluxated teeth, an unexpected finding was that the L.D.F. classification changed over time for 13 of the 26 teeth (50% of the sample). Seven teeth changed from L.D.F. Non-vital to L.D.F. Vital and of these teeth, six were followed up long enough for continued root growth to be seen on radiographic examination. Two L.D.F. Vital teeth became L.D.F. Non-vital, with pulpectomy confirming the diagnosis on both occasions. Finally, the remaining four teeth changed L.D.F. classification on two occasions; two from L.D.F. Vital to Non-vital to Vital (with continued root growth noted for one of

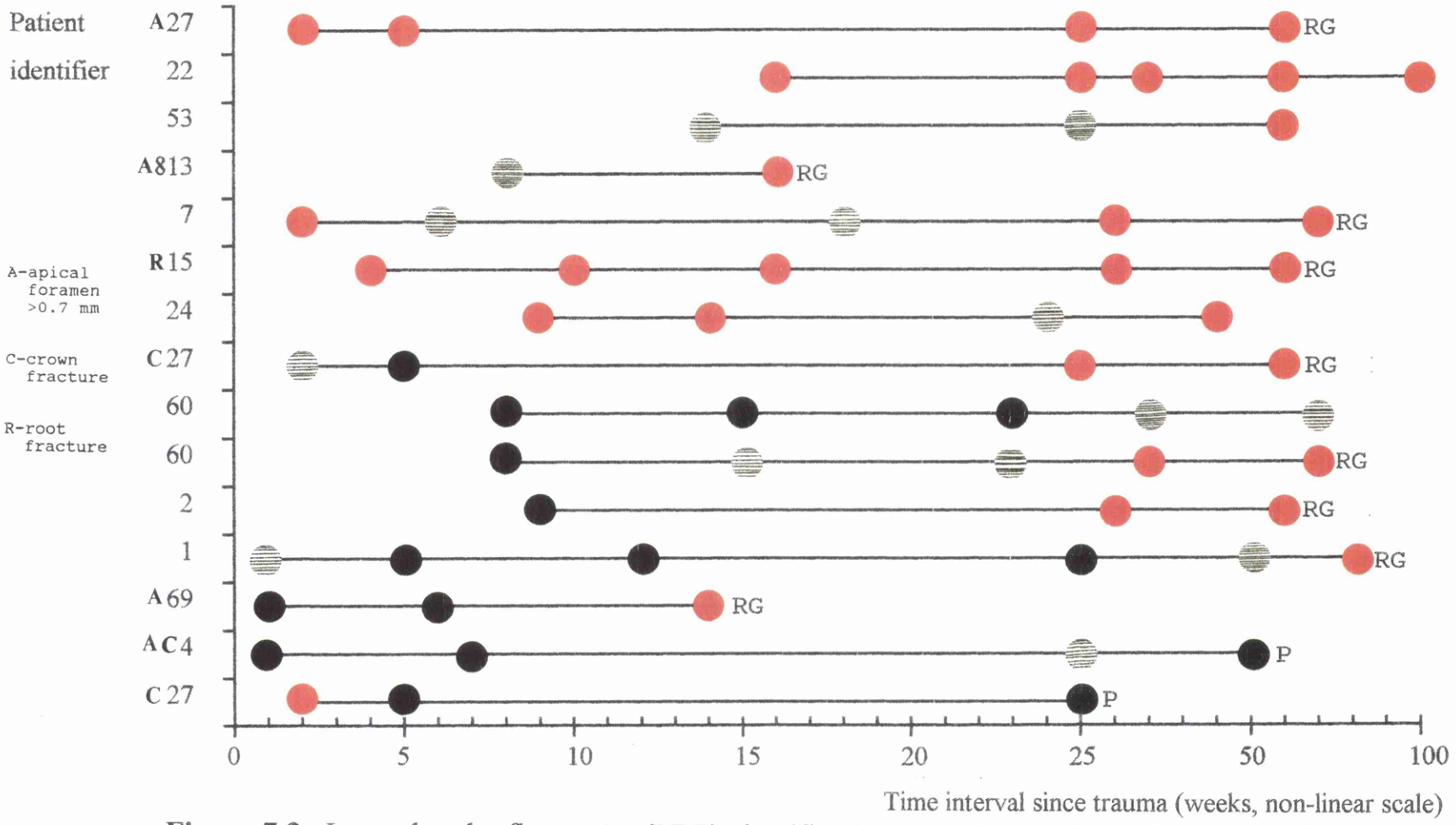


Figure 7.3 Laser doppler flowmetry (LDF) classification over time for luxated permanent incisors

-LDF Vital, RG -classification confirmed by continued root growth

-LDF Intermediate vitality

-LDF Non-vital, P -classification confirmed by pulpectomy

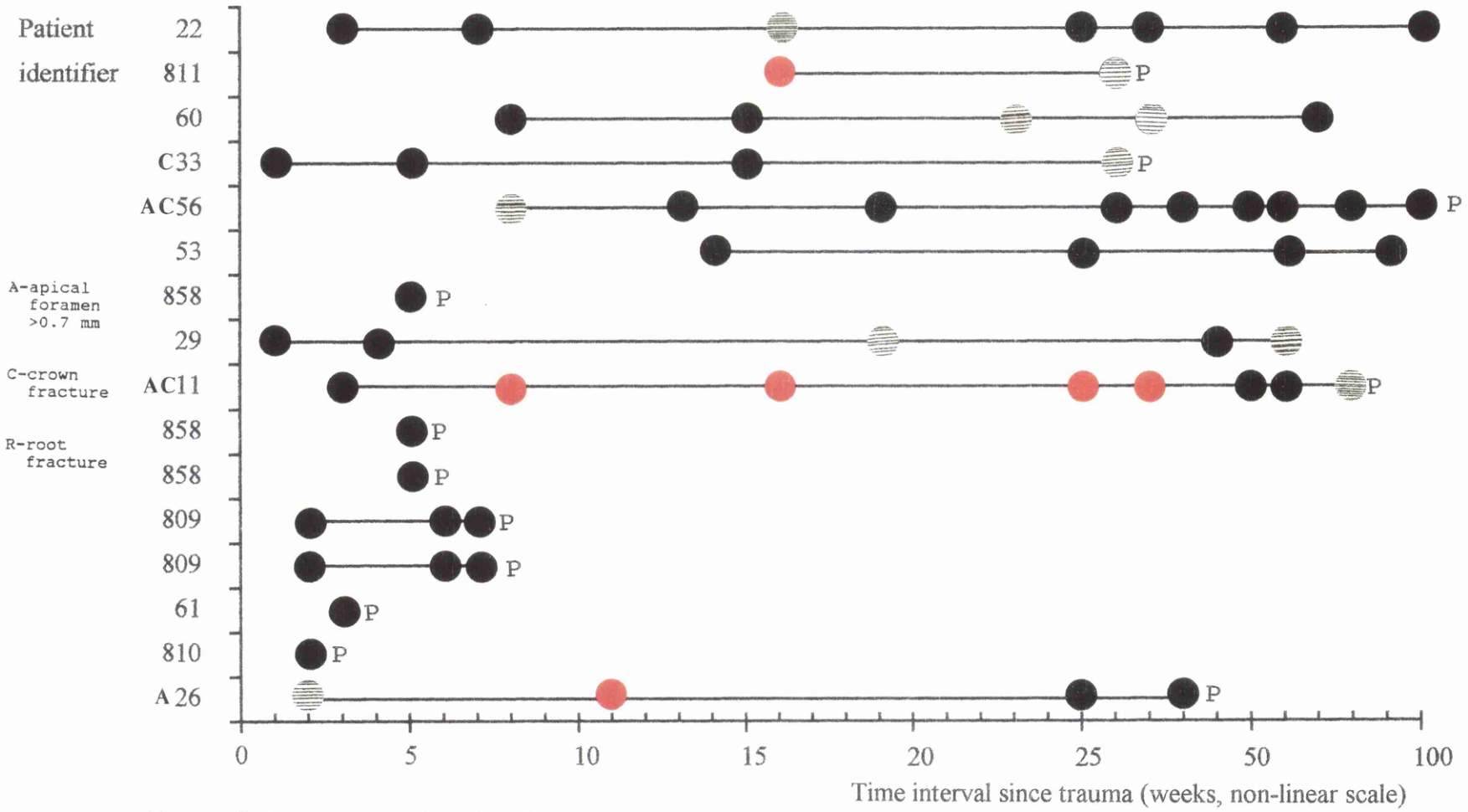


Figure 7.3(cont) Laser doppler flowmetry (LDF) classification over time for luxated permanent incisors

-LDF Vital, RG -classification confirmed by continued root growth

-LDF Intermediate vitality

-LDF Non-vital, P -classification confirmed by pulpectomy

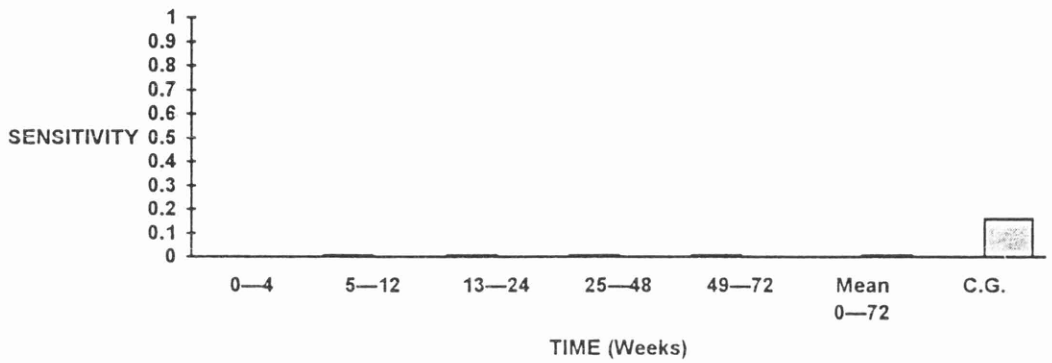
them), while the other two changed from L.D.F. Non-vital to Vital to Non-vital (with both confirmed non-vital by pulpectomy). The timings of these changes relative to the injury showed wide variation, as can be seen from Figure 7.3, and were noted between two and 50 weeks following injury.

The data for test sensitivities and specificities are shown in Tables 7.25-7.36. The values for mean sensitivities over all the time intervals were generally either the same as, or a little less than, the values obtained from the Comparison Group. Individual tests occasionally showed a slightly greater fall during certain time intervals; for example, the test of tenderness to percussion showed a fall in sensitivity of 0.35 compared with the comparison group at time intervals 13-24 weeks (sample size 10 teeth) and again at 49-72 weeks (sample size eight teeth). The sensitivity of the tests of crown colour and transillumination fell steadily over the time periods. However, the mean values for sensitivity over all the time periods for these two tests and for all the other tests including pulpal sensibility, never fell by more than 0.20 from the values obtained from the Comparison Group.

Excluding tests of pulpal sensibility, the specificities of the tests followed the same pattern, being generally the same or a little below the values obtained from the comparison group. With the exception of the test of transillumination, which showed a fall in specificity of 0.50 during the time period 0-4 weeks following trauma, no test showed a fall of more than 0.20 below the value obtained from the Comparison Group in any time interval. The mean values for all the tests (except tests of pulpal sensibility) over all the time intervals varied by only between +0.03 and -0.11 from the values obtained from the Comparison Group. However, the specificities of the tests of pulpal sensibility showed a different trend. The specificity of ethyl chloride fell by as much as 0.89 in some time intervals and that of electric pulp testing by as much as 0.96 from the values obtained from the Comparison Group, with the mean specificities over time showing a fall of 0.72 for ethyl chloride and 0.65 for electric pulp testing.

Table 7.25 History of pain; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.00 n=19	0.00 n=9	0.00 n=13	0.00 n=9	0.00 n=50	0.16 n=55
Specificity	N/A	1.00 n=5	1.00 n=8	1.00 n=8	1.00 n=10	1.00 n=31	0.99 n=84



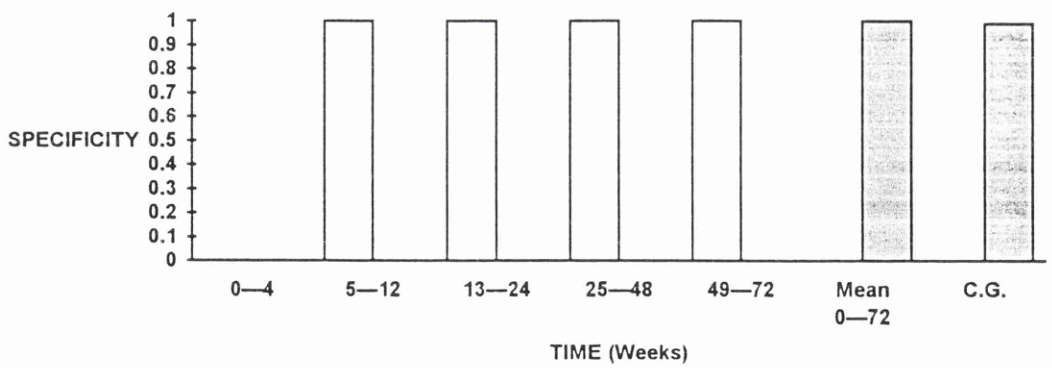
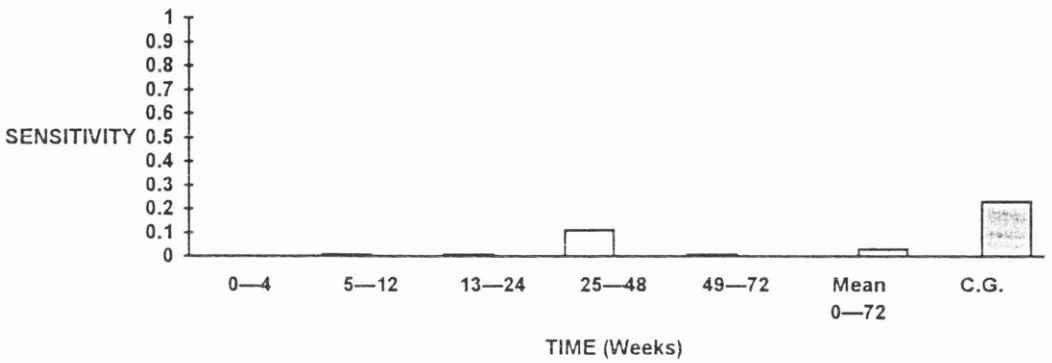


Table 7.26 Alveolar tenderness; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.00 n=8	0.00 n=7	0.11 n=9	0.00 n=8	0.03 n=32	0.23 n=43
Specificity	N/A	N/A	1.00 n=2	1.00 n=3	1.00 n=5	1.00 n=10	1.00 n=43



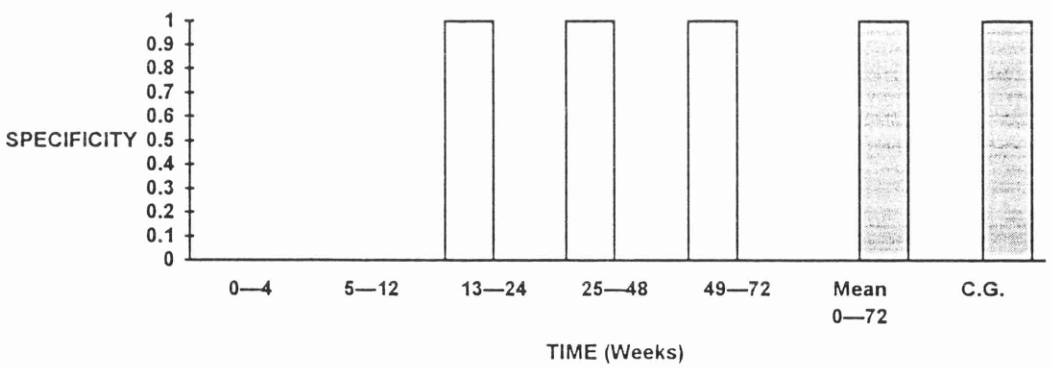
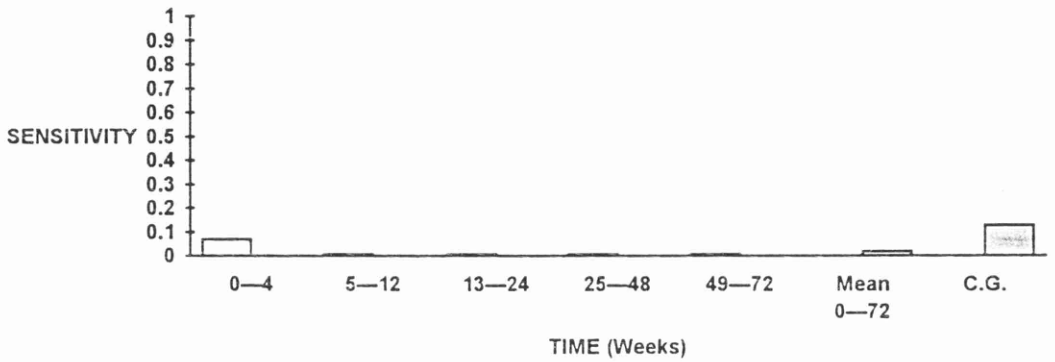


Table 7.27 Alveolar sinus; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.07 n=14	0.00 n=18	0.00 n=10	0.00 n=13	0.00 n=9	0.02 n=64	0.13 n=55
Specificity	1.00 n=4	1.00 n=5	1.00 n=8	1.00 n=8	1.00 n=10	1.00 n=35	1.00 n=83



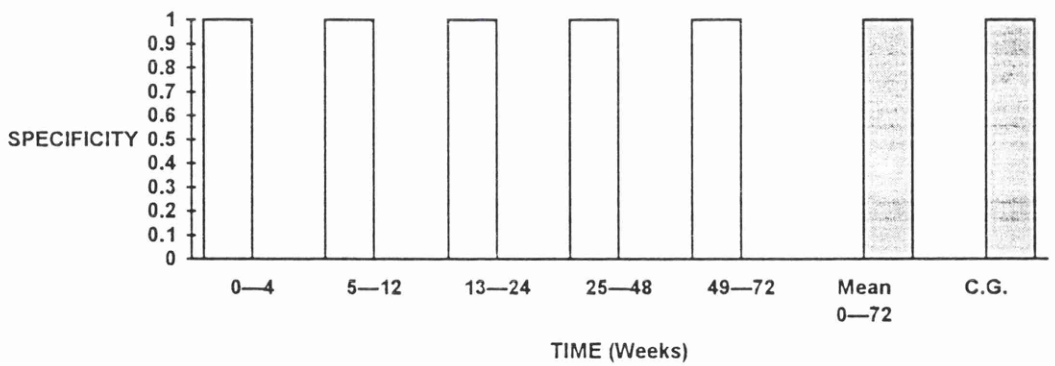
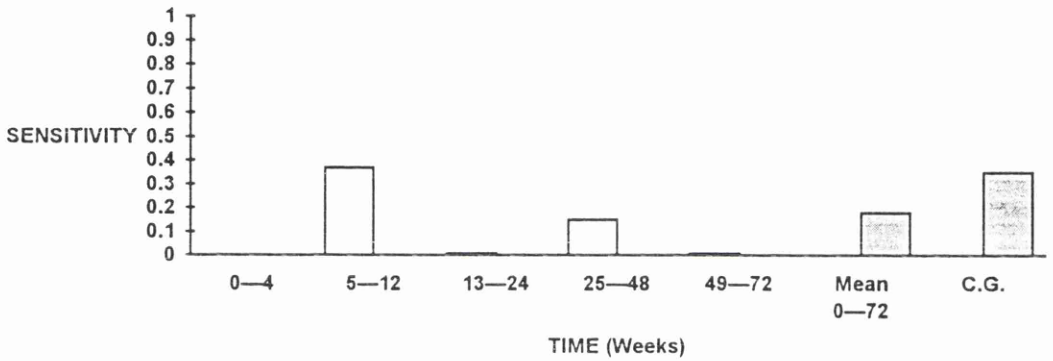


Table 7.28 Tenderness to percussion; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.37 n=19	0.00 n=10	0.15 n=13	0.00 n=8	0.18 n=50	0.35 n=54
Specificity	N/A	1.00 n=5	0.88 n=8	1.00 n=9	1.00 n=9	0.97 n=31	0.99 n=76



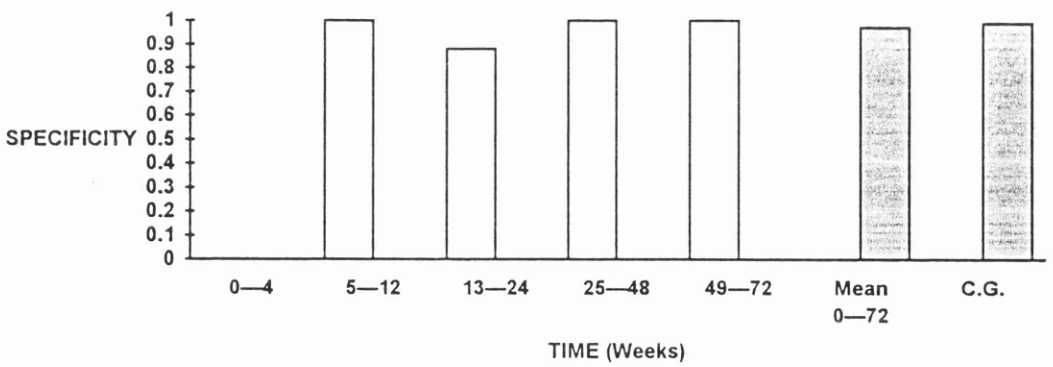
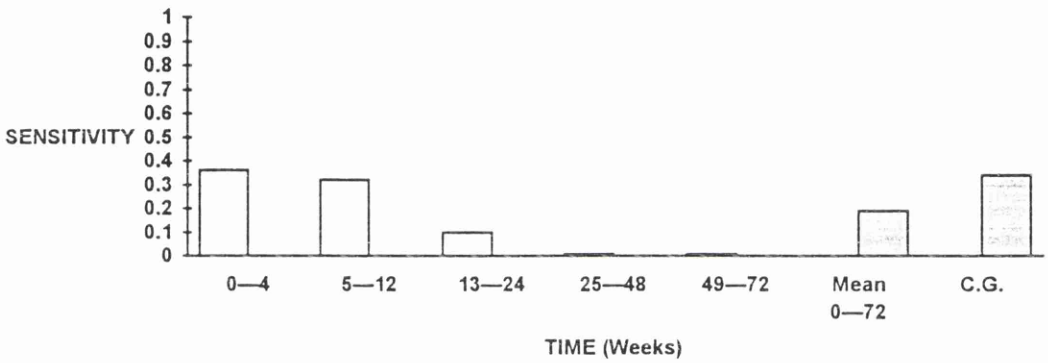


Table 7.29 Crown colour; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.36 n=14	0.32 n=19	0.10 n=10	0.00 n=13	0.00 n=9	0.19 n=65	0.34 n=53
Specificity	1.00 n=4	1.00 n=5	1.00 n=8	1.00 n=9	1.00 n=10	1.00 n=36	1.00 n=79



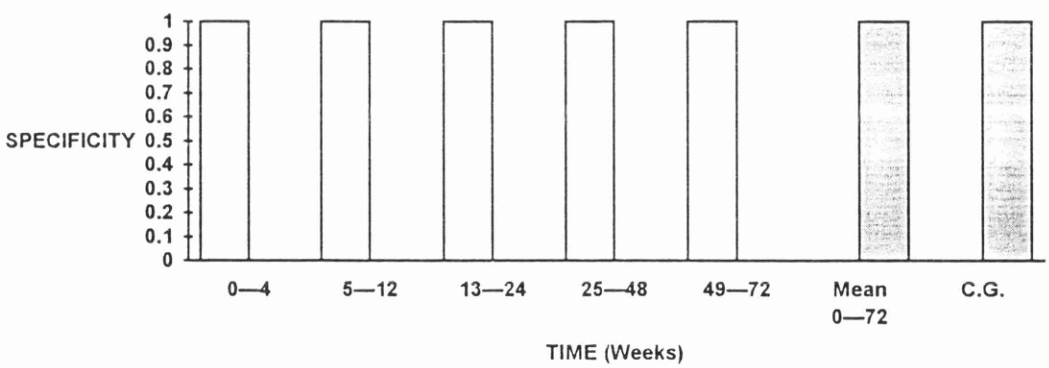
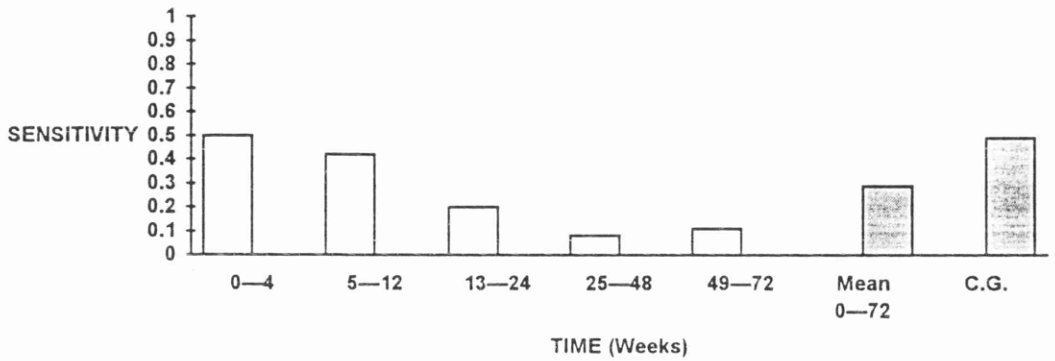


Table 7.30 Transillumination; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.50 n=14	0.42 n=19	0.20 n=10	0.08 n=13	0.11 n=9	0.29 n=65	0.49 n=51
Specificity	0.50 n=4	1.00 n=5	0.88 n=8	1.00 n=9	0.90 n=10	0.89 n=36	1.00 n=77



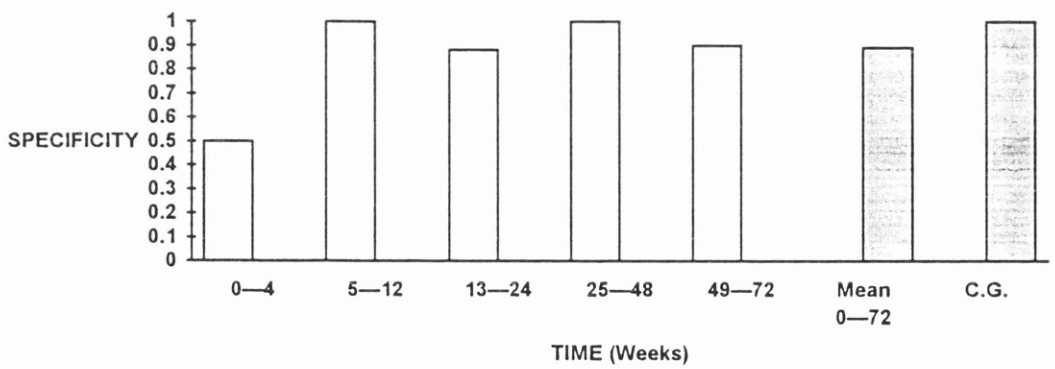
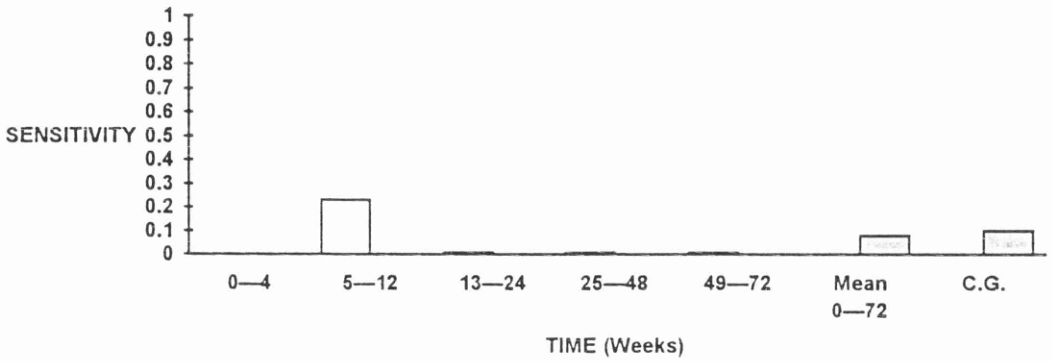


Table 7.31 Mobility; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	0.23 n=13	0.00 n=6	0.00 n=10	0.00 n=8	0.08 n=37	0.10 n=41
Specificity	N/A	1.00 n=1	1.00 n=3	0.80 n=5	1.00 n=7	0.94 n=16	1.00 n=50



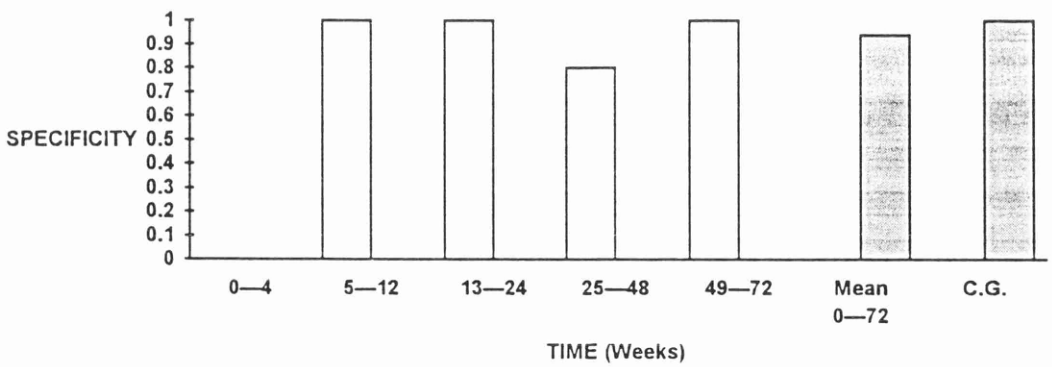
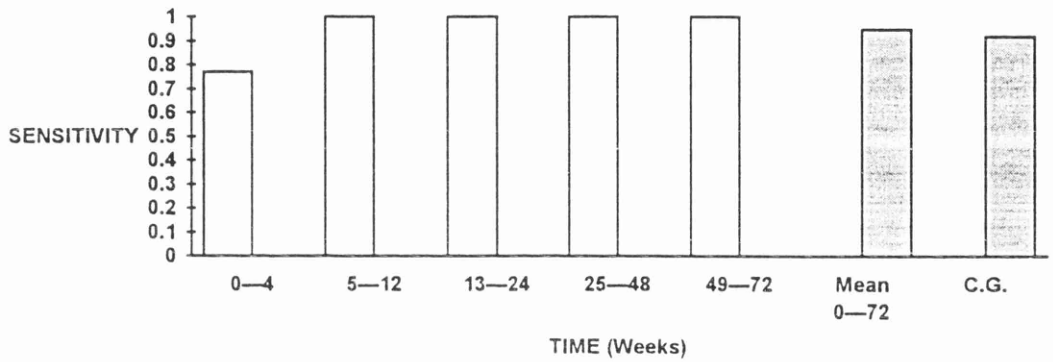


Table 7.32 Ethyl chloride; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.77 n=13	1.00 n=18	1.00 n=10	1.00 n=13	1.00 n=9	0.95 n=63	0.92 n=53
Specificity	0.00 n=4	0.20 n=5	0.29 n=7	0.11 n=9	0.20 n=10	0.17 n=35	0.89 n=81



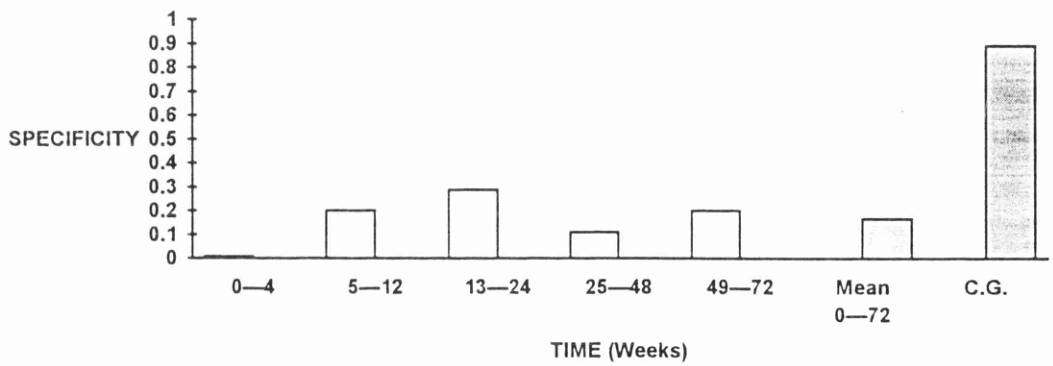
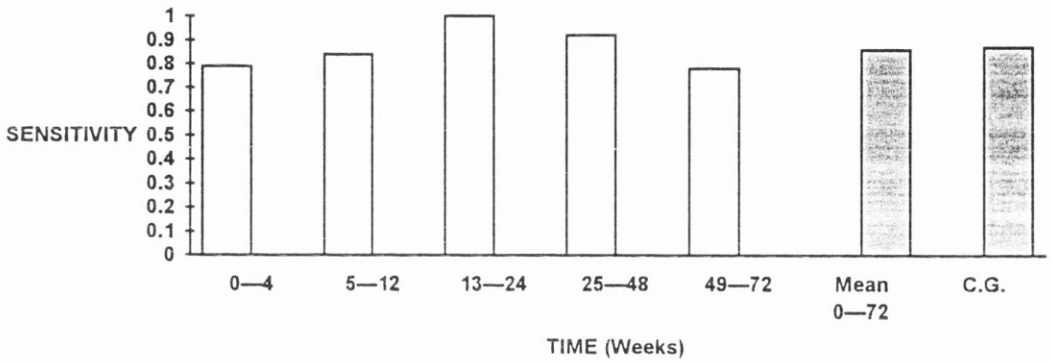


Table 7.33 Electric pulp test; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.79 n=14	0.84 n=19	1.00 n=10	0.92 n=13	0.78 n=9	0.86 n=65	0.87 n=53
Specificity	0.25 n=4	0.00 n=5	0.25 n=8	0.22 n=9	0.60 n=10	0.31 n=36	0.96 n=83



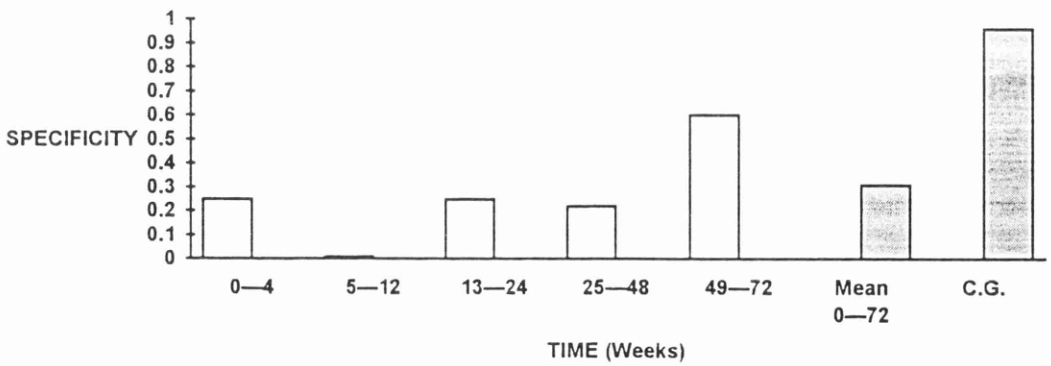
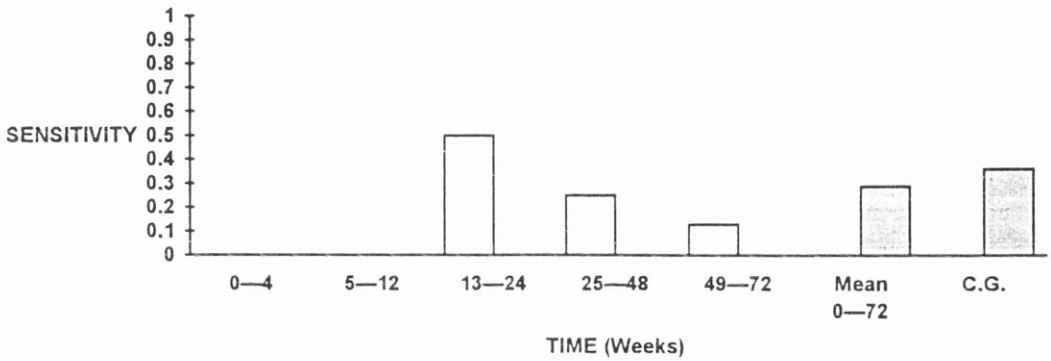


Table 7.34 Periapical radiolucency; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	N/A	N/A	0.50 n=8	0.25 n=8	0.13 n=8	0.29 n=24	0.36 n=47
Specificity	N/A	N/A	1.00 n=6	1.00 n=7	1.00 n=9	1.00 n=22	0.97 n=64



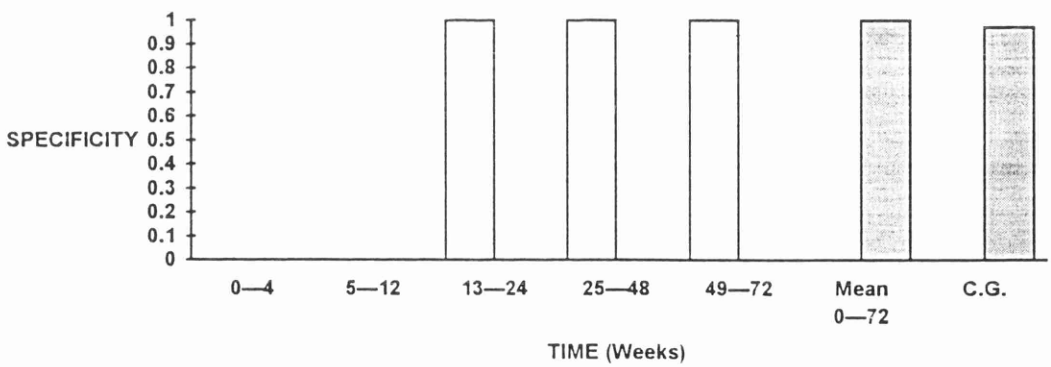
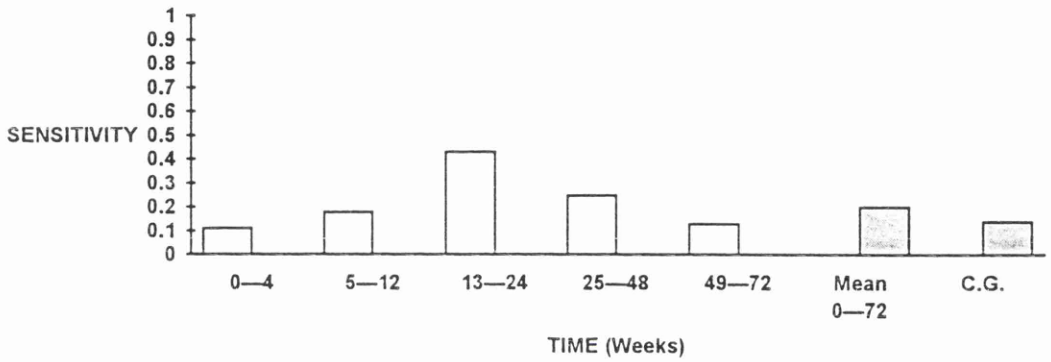


Table 7.35 Root apex resorption; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.11 n=9	0.18 n=17	0.43 n=7	0.25 n=8	0.13 n=8	0.20 n=49	0.14 n=53
Specificity	1.00 n=1	1.00 n=2	1.00 n=6	0.57 n=7	1.00 n=9	0.88 n=25	0.98 n=66



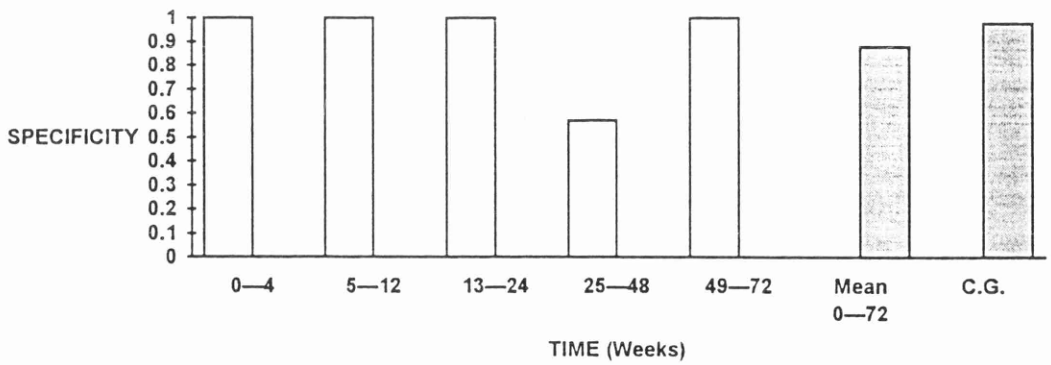
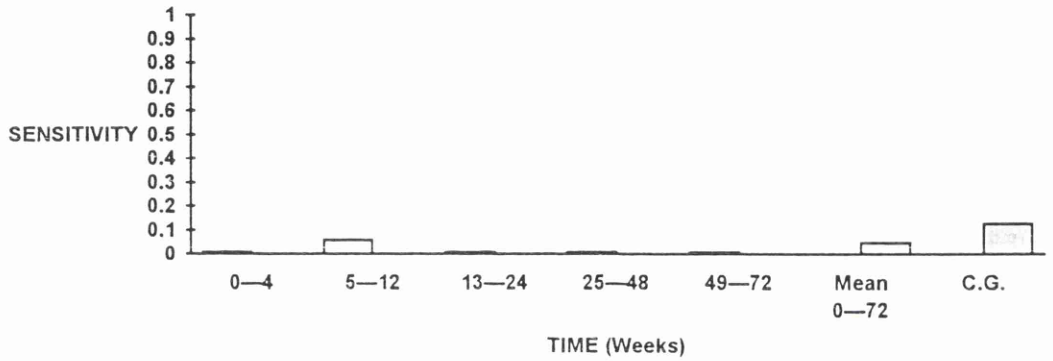
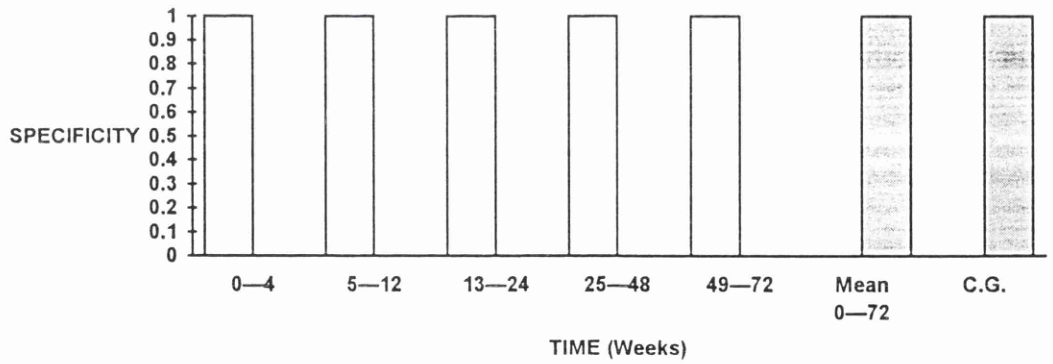


Table 7.36 External root resorption; diagnostic sensitivity and specificity when vitality testing luxated permanent anterior teeth. n=number of teeth in sample

Time post-trauma (weeks)	0—4	5—12	13—24	25—48	49—72	Mean 0—72	Comparison group (C.G.)
Sensitivity	0.00 n=9	0.06 n=17	0.00 n=7	0.00 n=8	0.00 n=8	0.05 n=49	0.13 n=53
Specificity	1.00 n=1	1.00 n=2	1.00 n=6	1.00 n=7	1.00 n=9	1.00 n=25	1.00 n=66





7.3.4 Avulsion injuries

Eight avulsed teeth (seven patients) were included in the study, and the L.D.F. classification over time is shown in Figure 7.4. Four of the patients presented more than 20 weeks following replantation. Three teeth were subject to immediate pulpectomy following a single L.D.F. Non-vital recording, with the pulpectomy confirming the diagnosis. The remaining five teeth became L.D.F. Vital, with continued root growth noted for three teeth. One tooth was L.D.F. Vital 16 days following replantation while another tooth did not become L.D.F. Vital for nearly a year and a half following replantation. Insufficient avulsed teeth were included in the study to allow valid analysis of the sensitivity and specificity of standard diagnostic tests.

7.4 DISCUSSION

The first aim of this chapter was to investigate the temporal relationship between dental trauma and pulpal vitality. There is a lack of clarity regarding the time interval following trauma after which the vitality of a tooth could be regarded as established. This may be due to the unreliability of existing methods of assessing pulpal status or to actual changes in pulpal status. The present study provides evidence that the pulpal status of teeth subjected to trauma is not necessarily established at the moment of trauma but may, in many cases, change over time following the injury. A change in pulpal status was noted in 45% of the sample of 49 subluxated and luxated teeth where two or more L.D.F. recordings were available. Of the 22 teeth where a change in L.D.F. classification was noted, this comprised revascularisation of a necrotic dental pulp for 12 teeth (55% of the sample), pulpal necrosis developing in a previously vital dental pulp in two teeth (9% of the sample) while for eight teeth (36% of the sample) the L.D.F. classification changed twice (the last classification being L.D.F. Vital for five teeth and L.D.F. Non-vital for three teeth). There was no obvious relationship between the time of trauma and the development of these changes, which were found to occur days, weeks or even a year

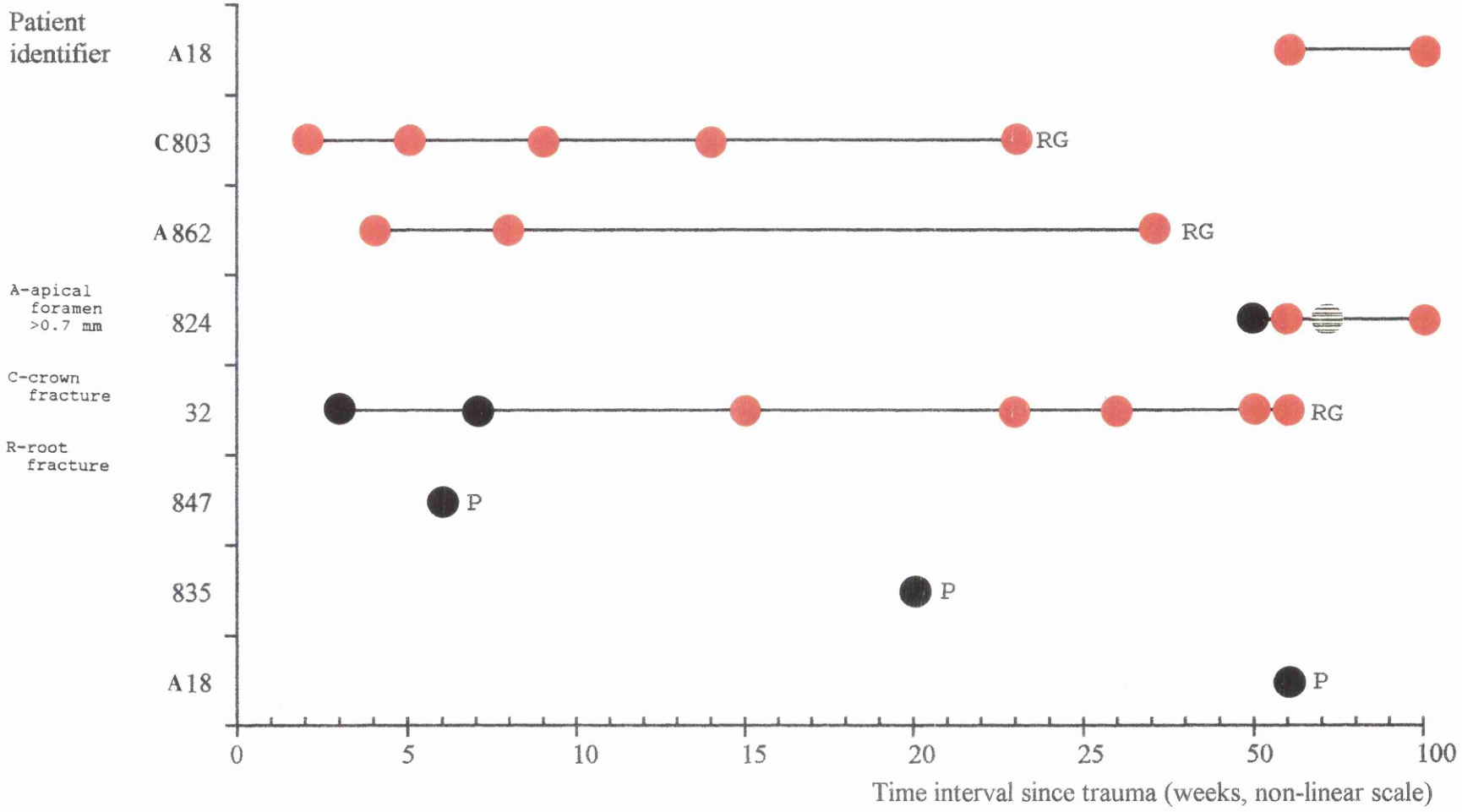


Figure 7.4 Laser doppler flowmetry (LDF) classification over time for replanted permanent incisors

-LDF Vital, RG -classification confirmed by continued root growth

-LDF Intermediate vitality

-LDF Non-vital, P -classification confirmed by pulpectomy

or more after injury. It is possible that, with prolonged follow up periods, changes in pulpal status might be noted after even longer time intervals.

Where a change in pulpal status was noted it was, in the majority of occasions, a revascularisation of a previously necrotic dental pulp and there were indications that the probability of this happening was not affected by the relative maturity of root development. Of the 20 luxated and subluxated teeth in the study where laser doppler flowmetry showed revascularisation, 15 teeth (75% of the sample) had root apices with a diameter <0.7 mm. This was the same proportion of teeth with root apices <0.7 mm as for the 49 luxated and subluxated teeth from which the sample was drawn.

The process of revascularisation can occur very rapidly, or may be delayed; an animal study by Skoglund *et al.* (1978), using microangiography, reported revascularisation of dental pulps by anastomotic union of blood vessels within four days. The finding of the present study that a replanted tooth had a coronal blood flow 16 days following replantation would tend to support the development of anastomotic union of pulpal blood vessels in humans. However, another replanted tooth revascularised more than a year following replantation and a review of the L.D.F. classifications for the traumatised teeth (Figures 7.1-7.4) shows no particular pattern to the timing of either pulpal revascularisation or the development of pulpal necrosis. The implications of these findings for the dental care of patients with traumatised teeth will be discussed in Chapter 8.

At present, it is only possible to speculate on the causation of changes in pulpal status although it seems likely that bacterial infection plays a central role (Section 2.3.5). The available evidence indicates that bacterial infection of a root canal is required to produce signs of radiographic periapical change (Section 2.8.2). Such a change, termed transient apical breakdown (Andreasen, 1986), was noted in four of the 20 non-vital subluxated and luxated teeth which subsequently revascularised (one of these cases is illustrated in Figure 8.1, Chapter 8). It would appear, therefore, that in these cases an existing infection of the dental pulp did not prevent revascularisation. Similarly, the five avulsed teeth which went on to revascularise must have had

infected, necrotic pulps at the time of replantation. It is, possibly, changes in the balance between bacterial infection and host resistance which causes the lability in pulpal status following trauma and further research in this area is indicated.

The second aim of Chapter 7 was to determine if, as might be anticipated, the sensitivity and specificity of standard pulpal diagnostic tests decreased if applied to a representative sample of traumatised teeth, as distinct from untraumatised vital teeth and non-vital traumatised teeth where at least two or more diagnostic tests indicated pulpal necrosis. It would seem that all standard pulpal diagnostic tests, with the exception of tests of pulpal sensibility, followed a similar trend of high mean specificities (range 0.88-1.0) and low mean sensitivities (range 0.00-0.46). They were, therefore, likely to correctly indicate that a vital pulp was vital although they were unlikely to correctly indicate that a non-vital pulp was necrotic. This balance of high specificities and low sensitivities, which means that vital pulps are unlikely to be subjected to an unnecessary pulpectomy but that necrotic pulps may go untreated, is probably to the patient's advantage (Section 2.4.2). There was a small decrease in reliability, particularly in mean sensitivities, when the standard tests (except pulpal sensibility tests) were applied to the sample of traumatised teeth as compared with the Comparison Group and this would support the need to always use these tests in combinations (Andreasen, 1988).

However, the tests of pulpal sensibility showed a different trend. While their mean sensitivities remained high for both traumatised teeth and the Comparison Group, their mean specificities showed the largest fall of all the tests when applied to subluxated teeth, and an even greater fall when applied to luxated teeth. This meant that, while a positive response to the sensibility testing of a vital traumatised tooth was likely to indicate vitality, no response did not reliably indicate a necrotic pulp. For example, of the nine vital luxated teeth tested between 25-48 weeks following trauma, only one responded to testing with ethyl chloride (specificity 0.11, Table 7.32). There was no indication from this study that the specificity of pulp testing of luxated teeth improved over time although, as has been discussed, sample sizes were small.

It is interesting to speculate on why sensibility testing was so unreliable, due particularly to the large fall in mean specificity, when assessing the vitality of traumatised teeth. There is general agreement, discussed further in Section 2.3, that pulpal neural tissue and pulpal vascular tissue may respond differently to traumatic injury. The finding that the mean specificities of sensibility tests decreased from concussion to subluxation injuries, and decreased again for luxation injuries indicates that the degree of disruption of the pulpal sensory neural supply is related to the severity of injury, and implies that neural tissue is more susceptible to traumatic injury than vascular tissue. There was also some indication that the diagnostic sensitivities of tests of pulpal sensibility were related to the severity of the dento-alveolar injury. Comparison of the mean sensitivities of pulp testing with ethyl chloride between subluxated teeth and luxated teeth (Tables 7.20 and 7.32) and of testing the same groups of teeth with electric pulp tests (Tables 7.21 and 7.33) show an increase in mean sensitivities of both tests with increasing severity of trauma. In fact, for the more severe of the two injuries, luxations, the mean sensitivities equalled those obtained from the Comparison Group.

The slightly lower mean sensitivities obtained from the subluxation group indicates that, on occasions, teeth were maintaining a neural response in the absence of a coronal blood supply. For example, five to twelve weeks following subluxation injury, four non-vital teeth out of a sample of 11 non-vital teeth still gave a positive response (sensitivity 0.64, Table 7.20) to testing with ethyl chloride. During the same time interval, none of the 18 non-vital luxated teeth tested with ethyl chloride gave a response (sensitivity 1.0, Table 7.32).

It would seem, therefore, that neural tissue is more susceptible than vascular tissue to dental trauma but that, on occasion, the neural tissue of teeth subjected to traumatic injury may remain functional in the absence of a coronal blood supply. This might be the consequence of an initially perfused traumatised dental pulp becoming necrotic over time due to pulpal inflammation or progressive infection (Section 2.3.4). This sequence of events might be possible following the relatively minor dento-alveolar injury of subluxation, but would seem unlikely following luxation injuries,

where complete rupture of the entire pulpal neurovascular supply would seem inevitable. The increased incidence of complete rupture of the neurovascular supply with luxation injuries would explain the increase in sensitivities of sensibility testing with increasing severity of trauma.

7.5 CONCLUSION

This study indicates that the vitality of traumatised teeth is not established at the moment of injury, but may change in the weeks, months or possibly years following the trauma. A change in the vitality of the dental pulp was noted in 45% of a sample of 49 subluxated and luxated teeth where two or more laser doppler flowmetry recordings were available, but there was no obvious pattern to the timing of these changes. For over half the cases the change comprised the revascularisation of a necrotic dental pulp and the likelihood of this process occurring seemed independent of the stage of root development of the tooth.

Standard diagnostic tests, excluding tests of pulpal sensibility, were unreliable in indicating the true status of a dental pulp in that they were unlikely to identify necrotic pulps (they had low sensitivities, generally <0.50). They were, however, very unlikely to indicate that a vital pulp was necrotic (they had high specificities, generally >0.90). This degree of unreliability was largely unaffected by whether the teeth being tested had or had not been subject to dental trauma.

Tests of pulpal sensibility showed a different trend. The tests were the most reliable of all the standard pulpal diagnostic tests when applied to either vital untraumatised teeth or to non-vital teeth where pulpal necrosis was indicated by two or more tests (high sensitivities and specificities, both values generally >0.90). However, when applied to vital traumatised teeth, the specificity of the tests fell in relationship to the severity of the dento-alveolar injury sustained. While for concussed teeth the specificities were similar to those obtained from vital untraumatised teeth (around 0.90), they fell by around 0.30 for vital subluxated teeth with a further fall of around 0.30 when applied to vital luxated teeth. Therefore, a negative response to pulpal sensibility testing of subluxated and luxated teeth should not be regarded as a reliable indication of pulpal necrosis.

The implications of these findings for the management of traumatised teeth are discussed in Chapter 8.

CHAPTER 8

A SUMMARY OF THE STUDY AND THE CLINICAL IMPLICATIONS OF THE FINDINGS

The clinical management of traumatised teeth is complicated by the unreliability of current methods of assessing dental pulp vitality. Studies by Gazelius *et al.* (1986 & 1988) indicated that laser doppler flowmetry had potential as a pulpal diagnostic aid, and a method of using the technique to discriminate between vital and non-vital dental pulps was developed during the studies reported in Chapters 4 and 5.

This method used a jig fabricated from a two-stage elastomeric impression material, in a modified stock impression tray, to hold the fibre-optic probe perpendicular to the tooth surface, between 2-3 mm from the gingival margin. It was found that signals of non-pulpal origin were an unavoidable component of laser doppler flowmetry of the dental pulp, but that by using this method they could be minimised to comprise only around 10% of the signal from a vital dental pulp. Recording methods reported in other studies of pulpal blood flow using laser doppler flowmetry were found to produce higher proportions of non-pulpal signals (up to 45% for one method), although this inherent problem with laser doppler flowmetry of the dental pulp was rarely acknowledged in the reports. The indications from this part of the study were that, due to the problems of non-pulpal signals from both non-pulpal blood flow and probe movement, the technical difficulties in developing a reliable, simplified, hand-held laser doppler flowmeter for pulpal assessment will be formidable.

Laser doppler flowmetry recordings taken from vital dental pulps, using the impression jig method, showed poor intra-patient repeatability. Optimum repeatability was obtained for the flux signal variable Mean Flux when the recording was repeated after only 10 minutes, but the prediction interval when repeating an individual recording was still -35% to +29% of the original recording. Application of

other recording variables, such as extending the review period to five weeks, reduced the repeatability, with the poorest repeatability found following exercise. At present, therefore, laser doppler flowmetry may have some use in recording changes in blood flow within the same recording session (although the non-linearity of the signal output should be considered (Section 3.2)), but would seem to have little potential in quantifying changes in pulpal blood flow over longer periods of time. Despite poor repeatability, flux signals from vital dental pulps were found to have two characteristics: a Mean Flux value $\geq$ 7 Perfusion Units and an amplitude of Slow Wave Vasomotion $\geq$ 1.6 Perfusion Units. These flux signal values formed the basis of the Laser Doppler Flowmetry classification criteria (Section 6.1), which were used to discriminate between signals from vital and non-vital dental pulps. Using these criteria, laser doppler flowmetry was found to have a sensitivity and a specificity of 1.0 when applied to a sample of 67 non-vital teeth and 84 vital teeth and was found to be more reliable than any other pulpal diagnostic test in current use.

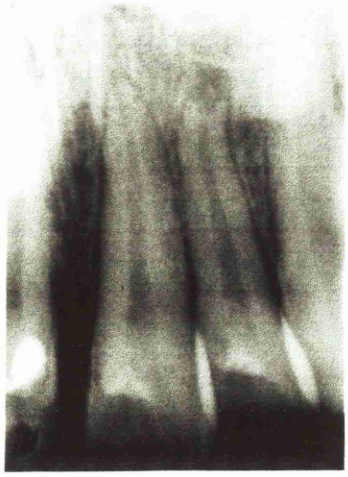
In general, standard pulpal diagnostic tests (with the exception of tests of pulpal sensibility) were found to have high specificities (range 0.88-1.00) but low sensitivities (range 0.00-0.49), whether applied to traumatised or untraumatised teeth. However, tests of pulpal sensibility (ethyl chloride (E.C.) and electric pulp test (E.P.T.)) were found to follow a different pattern. The generally high sensitivities (E.C. 0.92, E.P.T. 0.87) and high specificities (E.C. 0.89, E.P.T. 0.96) of the tests fell when the tests were applied to a representative sample of traumatised teeth. For subluxated teeth, both the mean sensitivity and mean specificity of pulpal sensibility tests fell by about 0.30 while for luxated teeth, the mean sensitivity returned to normal levels but the mean specificity fell even further, to 0.17 for ethyl chloride and 0.31 for the electric pulp test. The clinical implications are that tests of pulpal sensibility, unlike other pulp tests, show increased unreliability when applied to traumatised teeth, particularly a tendency to indicate false negatives for dental pulp vitality. For luxated teeth, vital dental pulps are more likely than not to fail to respond to sensibility testing.

Laser doppler flowmetry of the dental pulp requires a time-consuming and exacting technique for its use. It is, however, a reliable method for assessing pulpal status and could prove a valuable aid in improving the quality of care offered to the patient who has sustained dental trauma. However, any diagnostic technique, no matter how reliable, is only of value if the information provided is of use in clinical decision making. While laser doppler flowmetry gives a reliable diagnosis of pulpal status, uncertainty as to the appropriate clinical management of traumatised teeth remains.

The longitudinal study of traumatised teeth reported in Chapter 7 found that a change in pulpal status occurred in 45% of a sample of 49 subluxated and luxated teeth where there were two or more laser doppler flowmetry recordings available; for over half the cases the change was a revascularisation of a necrotic pulp. This figure might have been even higher if pulpectomy had not been carried out on several non-vital luxated teeth within a few weeks of injury, due to existing criteria for pulpectomy at that time (Figure 7.3 (cont.) and Section 7.2). The likelihood of revascularisation of a necrotic pulp was independent of whether the root development was complete or incomplete, and was noted over time intervals ranging from two weeks to over a year following injury. The finding that a necrotic dental pulp may revascularise, even in a tooth with a radiographically mature apex, means that a reliable diagnostic method for assessing pulp vitality does not always assist with the decision as whether or not to carry out pulpectomy.

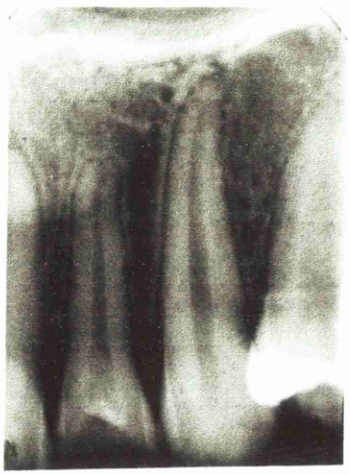
For example, Figure 8.1 shows radiographs of the maxillary left lateral incisor (22) of a 16-year-old female, A.M., which was subject to subluxation injury. A radiograph taken five weeks after injury shows no apparent pathological change. Laser doppler flowmetry at seven weeks post injury classified (22) as L.D.F. Vital, although the tooth did not responded to sensibility testing. When reviewed 11 weeks post injury, (22) was classified L.D.F. Non-vital, was non-responsive to sensibility testing, showed coronal discolouration and was tender to percussion. Contemporaneous radiographic examination showed (22) to have developed a periapical radiolucency and apical root resorption. As these were classic signs of

(22) 5 weeks post trauma
E.C., no response
E.P.T., no response
LDF Vital



(22) 11 weeks post trauma
E.C., no response
E.P.T., no response
LDF Non-vital

(22) 16 weeks post trauma
E.C., no response
E.P.T., responsive
LDF Intermediate vitality



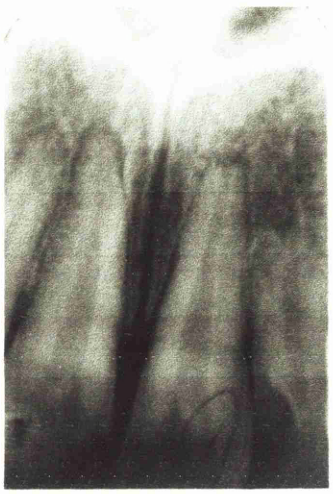
(22) 40 weeks post trauma
E.C., no response
E.P.T., no response
LDF Vital

(22) 92 weeks post trauma
Sensibility testing, no response

Figure 8.1 Periapical radiographs of maxillary left lateral incisor (22) in a 16 year old girl following subluxation injury (E.C.- ethyl chloride, E.P.T.- electric pulp test, LDF- laser doppler flowmetry diagnosis).

transient apical breakdown (Andreasen, 1986) pulpectomy of (22) was deferred. Reviewed 16 weeks post injury, (22) was classified L.D.F. Intermediate Vitality, was non-responsive to sensibility testing, showed coronal discolouration and was tender to percussion. Contemporaneous radiographic examination indicated that the periapical radiolucency affecting (22) had resolved. Reviewed 29 weeks post trauma, (22) was classified L.D.F. Intermediate Vitality, was non-responsive to sensibility testing but was no longer tender to percussion. Radiographic examination was deferred on this occasion. Reviewed 40 weeks post trauma, (22) was classified L.D.F. Vital, was non-responsive to sensibility testing and coronal discolouration remained. However, radiographic examination on the same occasion showed periapical healing. A radiograph taken a year later, 92 weeks post-trauma, showed continued narrowing of the root canal, although the tooth remained unresponsive to sensibility testing. On this occasion, deferring pulpectomy, despite some reasonably strong clinical indications, was in the patient's interest. However, deferring pulpectomy on traumatised teeth with necrotic pulps is not without risk for the patient.

Figure 8.2 shows a periapical radiograph of the maxillary left central incisor (21) of a 14-year-old female, L.C., 14 weeks following a luxation injury. The tooth was classified L.D.F. Non-vital at this time but as radiography indicated signs of transient apical breakdown, pulpectomy was deferred. The tooth was classified as L.D.F. Non-vital at further review periods of 29, 64 and 86 weeks post-trauma. At the 64 week review, radiography indicated periapical healing but at the 86 week review, radiography showed internal inflammatory resorption in the apical third of the root canal. Immediate pulpectomy confirmed that the coronal half of the root canal contained insensitive necrotic pulpal tissue, while brisk haemorrhage was encountered in the apical half of the canal, at the level of the resorption. It would seem probable that the revascularising front of tissue, advancing coronally along the root canal, encountered infection that could not be overcome in the middle third of the root canal, and that the resultant chronic inflammatory process resulted in internal root resorption. This patient had clearly been disadvantaged by deferring the pulpectomy for over a year following the trauma. Yet, for three patients in the study,



(21) 14 weeks post injury
Sensibility testing: non-responsive.
LDF Non-vital

(21) 86 weeks post injury
Sensibility testing: non-responsive.
LDF Non-vital

Figure 8.2 Periapical radiographs of maxillary left central incisor (21) in a 14 year old girl following extrusive luxation injury (LDF- laser doppler flowmetry diagnosis).

revascularisation of a necrotic dental pulp was not noted for over a year post-trauma. It is clear that reliable clinical criteria for pulpectomy of traumatised teeth still have not been developed and that further research in this area is required.

Andreasen (1989) stated that one of the greatest challenges facing dental traumatology was the diagnosis of pulpal and periodontal healing following traumatic injury. The present study has indicated that laser doppler flowmetry meets at least a part of that challenge, the reliable assessment of whether a traumatised dental pulp is vital or necrotic. However, an even greater challenge remains, and that is the reliable assessment of when the status of a traumatised pulp is established, and unlikely to change. This question will require further research before it can be answered and, therefore, contribute to informed, rational, evidence-based treatment decisions.

APPENDIX A

METHOD FOR RECORDING DENTAL PULP BLOOD FLOW USING THE PF2B LASER DOPPLER FLOWMETER.

INTRODUCTION

The method for recording dental pulp blood flow with the PF2b laser doppler flowmeter used in this study was as follows:

a) Pre-appointment

The setting up procedure started with sterilising the distal end of the L.D.F fibre optic probe in Cidex (Johnson and Johnson) for one hour before the patient attended. A spare probe was then fitted to the PF2b to allow it to be switched on for the minimum warm up period of one hour. Immediately prior to the patient attending, the PF2b was briefly switched off to change the probes, and a freshly mixed pellet of impression putty (Provil, Bayer) placed over the distal end of the sterilised probe. This acted both as a light stop and as a source of zero flux. The artifact filter on the PF2b was switched off, the waveband filter set at 4 KHz and the gain at the maximum of 100. The flux signal and Total Backscatter signal were output to a pen chart recorder (BBC Servigor) adjusted so that the flowmeters maximum output of 10 Volts gave a full scale deflection, equivalent to 100 Perfusion Units (P.U.), on the chart recorder. The flux signal was zeroed by adjusting the chart recorder and was only assessed if the Total Backscatter signal was above 20 P.U..

b) Patients appointment

The jig to hold the fibre optic probe was fabricated at the chairside, using a stock plastic lower dentate impression tray trimmed until only the six anterior teeth were included. This prevented patients occluding on the distal heels of the jig, producing artifactual signals. A single stage putty and wash elastomeric impression

(Provil, Bayer) was taken, ensuring a minimum labial thickness of 5 mm of impression material, and clear definition of gingival margins. Holes were prepared, using an 0.016 mm diameter slow speed rosehead bur in a latch grip right angle handpiece, to hold the probe perpendicular to the tooth surface. The gingival perimeter of the hole was between 2 mm and 3 mm from the gingival margin. In cases of gingival recession, the distance was measured from the amelo-cemental junction. The fit surface of the probe hole was countersunk with an 0.018 mm diameter rosehead bur to avoid tags of impression material obstructing the tip of the probe. The probe was then inserted until the distal end protruded through the fit surface of the impression, and the residual swarf removed with a blast of air from a triple syringe. The probe was then pulled back until the probe tip was 0.5 mm below the surface of the impression. This prevented the probe tip becoming scratched by the tooth surface on insertion, a problem which seriously affected the optical properties of the probe, leading to difficulties achieving the minimum Total Backscatter signal level of 20 P.U.. A small drop of clear water based lubricating jelly was then applied to the probe tip immediately prior to inserting the impression in the patient's mouth. Recordings of pulpal blood flow were taken with the patient supine and rested. The pen recorder chart speed was set at 3 cm per minute for three minutes and then at 60 cm per minute for about 20 seconds in order to observe the cardiac pulse signal.

c) Post appointment

The PF2b was switched off, the probe removed and the distal end disinfected in Cidex (Johnson and Johnson) for ten minutes before being rinsed, dried and packed away. The chart recording was analysed as described in Chapter 3.

APPENDIX B

TRANSCRIPT OF CONFERENCE PRESENTATION PUBLISHED IN THE INTERNATIONAL ENDODONTIC JOURNAL (1993)23:18-19

A CLINICAL COMPARISON OF TWO LASER DOPPLER FLOWMETERS WITH DIFFERENT WAVELENGTHS IN AN INVESTIGATION OF PULPAL BLOOD FLOW

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An investigation was undertaken to determine the relative performance of two laser doppler flowmeters in differentiating between clinically vital and non-vital anterior teeth. One instrument utilised a laser source of wavelength 633 nm (1) and the other a wavelength of 810 nm (2). Recordings were taken with both instruments from six clinically non-vital teeth from five patients. An elastomeric impression material was used to hold the fibre optic probes at 2 mm and at 4 mm from the gingival margin. Results are expressed as a mean ratio (+ or - Standard Deviation) of the flux from the non-vital tooth to the flux recorded from the vital control tooth from the same patient. The 633 nm wavelength flowmeter gave ratios of 0.19 (+ or - 0.08) at 2 mm, and 0.15 (+ or - 0.1) at 4 mm from the gingival margin. The 810 nm wavelength flowmeter gave ratios of 0.67 (+ or - 0.25) at 2 mm and 0.42 (+ or - 0.12) at 4 mm from the gingival margin. The performance of the 810 nm wavelength flowmeter was improved when recordings were taken at 4 mm rather than 2 mm from the gingival margin.

A further seven non-vital teeth were selected from a different group of six patients and recordings taken at 2 mm from the gingival margin using the 633 nm wavelength flowmeter and at 4 mm from the gingival margin using the 810 nm wavelength flowmeter. The ratio recorded by the 633 nm wavelength flowmeter was 0.15 (+ or - 0.08), with the 810 nm wavelength flowmeter recording a ratio of 0.44

(+ or - 0.3). Both instruments recorded pulse synchronicity and slow wave vasomotion in all control teeth. Both these parameters were absent from all the non-vital teeth recordings made with the 633 nm wavelength instrument but were absent in only one of the non-vital recordings made with the 810 nm wavelength instrument.

This pilot study indicates that a laser source of 633 nm improves the performance of the instrument in differentiating between clinically vital and non-vital teeth, compared with an 810 nm source. However, further work is required to show whether this improvement is due to wavelength or fibre optic design.

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(2) MBF3, Kindly loaned by Moor Instruments, Millwey, Axminster, Devon.

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