

## Review

## Bovine teeth as substitute for human teeth in dental research: a review of literature

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(Received 25 April and accepted 15 June 2011)

**Abstract:** The aim of this paper was to review *in vitro* and *in situ* studies that directly compared the use of bovine teeth as a substitute for human teeth in dental experiments. A PubMed search was conducted for papers published from 1953 to December 30, 2010 using the following keywords: “human bovine enamel” or “human bovine dentin” or “human bovine teeth”. The abstracts of the studies resulting from the keyword search were read, and all papers that compared human and bovine teeth were fully read. Only original articles written in English and directly comparing human and bovine substrates were included in the review. The search was supplemented by manual searches of the reference lists from each identified paper. Out of 76 studies initially selected, 68 fulfilled the selection criteria for inclusion. The studies covered seven categories: dental morphology, chemical composition, physical properties, dental caries, dental erosion/abrasion, bonding/adhesive strength, and marginal microleakage. Inconsistent data exist regarding whether bovine teeth can be considered an appropriate substitute for human teeth in dental research. Morphological, chemical composition and physical property differences between the two substrates must be considered when interpreting results obtained from any experiment using bovine tooth substrate. (J Oral Sci 53, 273-282, 2011)

**Keywords:** human teeth; bovine teeth; dental caries; dental erosion; bonding strength; microleakage.

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### Introduction

Specimens generated from human teeth are preferred for *in vitro* and *in situ* dental research because they allow for testing of the study hypothesis in a more clinically relevant substrate. However, some disadvantages and limitations with the use of human teeth exist (1). They are often difficult to obtain in sufficient quantity and with adequate quality, since many are extracted due to extensive caries lesions and other defects (1). It can also be challenging to control the source and age of the collected human teeth, which may lead to larger variations in the outcome measures of the study (2). Furthermore, the relatively small and curved surface area of human teeth may also be a limitation for specific tests requiring flat surfaces of uniform thickness (2). Finally, awareness of the infection hazard (3) and ethical issues (4) have increased. Therefore, alternative substrates have been proposed and used in dental research.

Several types of non-human teeth have been utilized as substrates for *in vitro* and *in situ* dental experiments. Common examples are primate (5), bovine (6), swine (7,8), equine (9), and shark teeth (10). However, bovine teeth have been the most widely used substitute for human teeth in dental studies and their use has dramatically increased in the last 30 years. Bovine teeth are easy to obtain in large quantities, in good condition and with a more uniform composition than that of human teeth. Furthermore, bovine teeth have a relatively large flat surface, and do not have caries lesions and other defects

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that might affect outcomes (1). Although bovine teeth have commonly been used, some concerns about the application of data obtained from bovine to human teeth have been raised, as their chemistry and structure are not identical (11-13). Studies comparing both substrates have been performed and published in different fields of dental research. The aim of this review was to identify these studies, including both *in vitro* and *in situ*, to compare their results, and to verify the validity of using bovine teeth as a substitute for human teeth in dental experiments.

### Search strategy

PubMed was searched for papers published from 1953 to December 30, 2010. The search keywords were: "human bovine enamel" or "human bovine dentin" or "human bovine teeth". The abstracts of the studies resulted from the keyword search were read. Only original articles written in English, directly comparing between human and bovine substrates were included in the review and fully read. The search was supplemented by manual searches of the reference lists from each identified paper. Interim reports, abstracts, letters, short communications, reviews, and chapters in textbooks were excluded from the final review.

### Search results

Out of 1073 abstracts originally identified, 76 were initially selected. Eight papers were excluded after evaluation of the full paper (14-21), as they indirectly compared human and bovine teeth. Therefore, 68 papers were included in the final review. The studies covered seven main categories: morphology (5 studies) (22-26), chemical composition (11 studies) (27-37), physical properties (12 studies) (38-49), dental caries (7 studies) (6,9,50-54), dental erosion/abrasion (10 studies) (55-64), bonding/adhesive strength (17 studies) (65-81), and marginal microleakage (7 studies, with one duplicate between bonding strength and marginal microleakage) (7,78,82-86).

### Micro-morphology studies

Five studies were reviewed (22-26). All of the studies used scanning electron microscopy (SEM). Arends and Jongbloed (22) studied the average diameter of enamel crystallites in human and bovine teeth. Larger diameters were found for bovine crystallites, with a bovine: human ratio of 1.6:1. Schilke et al. (23) showed no significant differences in the number and mean of dentinal tubules between bovine coronal dentin and the dentin of human primary and permanent molars. Camargo et al. (24)

found a significantly higher number of dentin tubules in bovine teeth compared to human teeth. However, the diameter of tubules was the same in both types of specimens. Camargo et al. (25) compared the superficial morphology of bovine and human sclerotic dentin. No significant difference was found in the number of open dentin tubules in either species. In contrast, Lopes et al. (26) compared the tubular dimensions and distribution of human and bovine dentin in superficial, middle and deep dentin regions. The authors found that the number of tubules per square millimeter, regardless of the region, was significantly higher in human dentin than in bovine dentin.

### Chemical composition studies

Eleven studies were reviewed (27-37). Bisaz et al. (27) showed detectable amounts of inorganic pyrophosphate in enamel and dentin of both human and bovine teeth. Feagin et al. (28) showed that the Ca/P ratio of the mineral removed from the enamel surfaces during demineralization, as well as the remineralization characteristics, were the same in both human and bovine enamel. Davidson et al. (29) found that the calcium content by weight of bovine and human tooth enamel was 37.9% and 36.8% respectively, and the calcium distribution was more homogenous in bovine enamel compared to human enamel. Mellberg and Loertscher (30) showed that fluoride uptake by sound permanent human and primary bovine enamel was different. Gwinnett et al. (31) found no significant difference in fluoride uptake between etched human and bovine enamel. Sydney-Zax et al. (32) showed no significant difference in carbonate content between human and bovine enamel at different stages of their development. However, the study compared primary human teeth to bovine teeth and did not clarify if primary or permanent bovine teeth were used. Camargo et al. (33) compared the calcium ion release and the pH of storage solutions between human and bovine teeth after the application of calcium hydroxide pastes with four different paste vehicles. The study revealed no significant difference between bovine and human teeth in the pH measurement, but bovine teeth showed significantly higher calcium ion release compared to human teeth.

Fincham (34) found that enamel matrix proteins isolated from developing human incisors showed overall amino acid composition similar to bovine developing enamel. However, some differences in electrophoretic patterns of enamel matrix between the two species were noted. These results were confirmed later using chromatographic, electrophoretic, and amino acid analyses (35). On the other hand, major differences between protein

contents of primary bovine and human enamel were found in another study (36). Jameson et al. (37) showed that the magnitude of water loss was significantly greater for bovine than for human dentin. However, rehydration of 99% of the weight of bovine dentin and 100% of the weight of human dentin was achieved in the study.

### Physical properties studies

Twelve studies were reviewed (38-49). Putt et al. (38) found no significant difference in the polishing degree rank of seven abrasives for human and bovine enamel. Spitzer and Bosch (39) found that the refractive indices were not significantly different between human and bovine enamel. However, the absorption peak at 270 nm in the bovine enamel spectrum was three times higher than that in the spectrum of human enamel. The same authors indicated no substantial differences between the luminescence of human and bovine enamel (40). On the other hand, Zijp and ten Bosch (41) revealed differences between human and bovine dentin both in scattering and anisotropy. Yu et al. (42) showed that mean translucency values of 1 mm thick bovine enamel, bovine dentin,

human enamel and human dentin were 14.7, 15.2, 18.7, and 16.4, respectively.

Reeh et al. (43) compared the effect of five different lubrication regimens *in vitro* between human and bovine enamel. The authors found high correlations between different lubricants on the two enamel substrates. Sano et al. (44) found no significant difference between mineralized or demineralized dentin of human and bovine teeth in either ultimate tensile strength or modulus of elasticity. Schmalz et al. (45) observed no significant difference between human and bovine dentin in hydraulic conductance and diffusional water flux. However, the authors found that the variability of the permeability data for bovine dentin was about half that of human dentin. Soares et al. (46) observed no significant difference in fracture strength of composite partial denture that is fixed to either human or bovine teeth, regardless of the use of fiberglass reinforcement or not.

Tanaka et al. (47) found that the radiographic density of bovine enamel and coronal dentin was significantly higher than that of human enamel and coronal dentin, respectively. However, no significant difference in the

Table 1 Summary of studies compared directly between human and bovine teeth in dental caries

| Author(s)                            | Pertinent aim of the study  | Type of study                | Enamel/dentin | Root/crown | Technique(s) used   | Pertinent outcome   |
|--------------------------------------|---|------------------------------|---------------|------------|---|---|
| Anderson et al. (50) (1998)          | Compare the local integrated mineral loss through small areas of the natural surface of human and bovine teeth during demineralization.   | <i>In Vivo</i>               | Enamel        | Crown      | Scanning microradiograph  | No clear distinction in mineral loss was found between human and bovine enamel.   |
| Edmunds et al. (9) (1988)            | Compare artificial caries lesions created with bacterial challenge or acidic gel between human and bovine teeth.  | <i>In vitro</i>              | Enamel        | Crown      | Polarized light microscopy<br>SEM   | The depth of caries lesion in human enamel was half of that in bovine for both types of lesion, similar depth ratio was seen in SEM, and structure of the lesion was the same in human and bovine enamel. |
| Featherstone and Mellberg (6) (1981) | Compare the rates of artificial caries lesion production in permanent human and bovine teeth using two different caries production systems.                                     | <i>In vitro</i>              | Enamel        | Crown      | Dye imbibitions<br>Microhardness tester   | Caries lesion progressed 2 times faster in bovine enamel compared to human enamel.  |
| Hara et al. (52) (2003)              | Compare bovine and human teeth in caries progression, inhibition and biofilm composition.   | <i>In situ</i>               | Dentin        | Root       | Microbiological analysis<br>Cross sectional microhardness<br>Polarized light microscopy | No significant difference was found between human and bovine dentin in caries progression, inhibition and composition of biofilm formed.  |
| Kielbassa et al. (53) (2006)         | Evaluate the effects of irradiation and surface condition on remineralization of dematerialized human and bovine enamel.  | <i>In situ/<br/>In vitro</i> | Enamel        | Crown      | Transverse microradiography   | Abrasion hampered demineralization in irradiated bovine specimens but not human specimens.  |
| Souza-Gabrial et al. (51) (2010)     | Compare the ability of CO <sub>2</sub> laser irradiation, fluoride varnish, and fluoride gel to inhibit the progression of artificial caries lesion in human and bovine enamel. | <i>In vitro</i>              | Enamel        | Crown      | Cross sectional microhardness<br>SEM  | Significantly higher microhardness in bovine enamel compared to human enamel.<br>SEM showed no significant qualitative difference between bovine and human enamel.  |
| Teranaka et al. (54) (1986)          | Compare the protein contents of artificially created bovine shallow caries lesions exposed to the mouth, and natural arrested human caries lesion.                              | <i>In situ/<br/>In vitro</i> | Enamel        | Crown      | Beckman 121-M amino-acid analyzer   | Incorporated organic material in bovine early caries lesion changed with time from having components dissimilar to natural human arrested caries lesion to ones similar to it.                            |

radiographic density was found between bovine and human radicular dentin. Fonseca et al. (48) showed no significant difference in radiodensity between human and bovine enamel or dentin. Fonseca et al. (49) compared the radiodensity and hardness of human and bovine

enamel and dentin of varying age. The study found that radiodensity was similar within enamel groups of human and bovine teeth. However, bovine dentin presented higher radiodensity than human regardless of age of the bovine teeth. Furthermore, Knoop hardness was similar

Table 2 Summary of studies compared directly between human and bovine teeth in dental erosion/abrasion

| Authors                       | Pertinent aim of the study  | Type of study          | Type of tooth     | Enamel/dentin    | Root/crown    | Technique used  | Pertinent outcome  |
|-------------------------------|---|------------------------|-------------------|------------------|---------------|---|--|
| Amaechi et al. (60) (1999)    | Determine the influence of enamel type on the development and progression of dental erosion.  | <i>In vitro</i>        | Permanent         | Enamel           | Crown         | Microradiography  | Significant lower mineral loss and lesion depth were observed in human permanent enamel and human primary enamel compared to bovine permanent enamel.  |
| Attin et al. (59) (2007)      | Compare the tooth surface loss of human permanent and primary teeth with that of bovine permanent and primary teeth as induced by toothbrush abrasion, erosion and the combination of erosion and abrasion. | <i>In vitro</i>        | Permanent Primary | Enamel           | Crown         | Profilometry  | No significantly different in enamel loss between the different substrates after abrasion. Enamel loss of the human primary and permanent teeth was significantly lower than that in bovine primary and permanent teeth respectively, after both erosion and erosion-abrasion challenges.  |
| Imfield (62) (2001)           | Determined the mechanical effects of manual toothbrush on standard abrasive on human and bovine samples.  | <i>In vitro</i>        | Permanent         | Dentin           | Root          | Relative dentin abrasion<br>Surface roughness (profilometry)<br>Cleaning effect | No significant difference was observed in abrasion or surface roughening between human and bovine dentin. Staining was easier to remove from human dentin compared to bovine.  |
| Meurman and Frank (58) (1991) | Compare dental erosion process qualitatively between human and bovine prismatic enamel that were immersed in acidic soft drinks for various time intervals.   | <i>In vitro</i>        | Permanent         | Enamel           | Crown         | SEM   | No difference was noticed in the progression of erosion and in the surface ultrastructure of erosive lesions between human and bovine substrates.  |
| Rios et al. (55) (2006)       | Compare quantitatively the behavior of bovine and human enamel substrates in abrasive lesions.  | <i>In situ/ex vivo</i> | Permanent         | Enamel           | Crown         | Surface microhardness<br>Wear profile   | The bovine enamel showed significantly less microhardness value and higher wear as compared to human enamel.   |
| Rios et al. (56) (2008)       | Evaluate the effect of stimulated saliva on the enamel surface of bovine and human substrates submitted to erosion followed by brushing abrasion immediately or after one hour.                             | <i>In situ</i>         | Permanent         | Enamel           | Crown         | SEM   | No differences between human and bovine enamel was observed.   |
| Turssi et al. (57) (2010)     | Compare human and bovine substrates in two dental erosion models.   | <i>In situ</i>         | Permanent         | Enamel<br>Dentin | Crown<br>Root | Surface microhardness   | No significant difference in microhardness between human and bovine enamel. Microhardness was significantly higher in human root dentin compared to bovine root dentin.  |
| Wegehaupt et al. (64) (2008)  | Compare the dentin wear caused by erosion-abrasion of permanent and primary human and bovine teeth.   | <i>In vitro</i>        | Permanent Primary | Dentin           | Crown         | Surface profilometry  | No significant difference in dentin wear induced by abrasion between primary and permanent human and bovine dentin respectively. Dentin wear induced by erosion was significantly higher for permanent and primary human than for permanent and primary bovine dentin respectively. No significant difference in dentin wear induced by abrasion-erosion between permanent human and bovine dentin. Dentin wear induced by abrasion-erosion was significantly higher for primary human dentin compared to primary bovine dentin. |
| Wegehaupt et al. (63) (2010)  | Compare the tooth surface loss of human and bovine dentin due to tooth brushing with different relative dentin abrasivity toothpastes.  | <i>In vitro</i>        | Permanent         | Dentin           | Root          | Contact profilometry  | No significant differences in the dentin wear between human and bovine root dentin.  |
| White et al. (61) (2010)      | Compare human and bovine erosion over acid-exposure times of two seconds to one hour.   | <i>In vitro</i>        | Permanent         | Enamel           | Crown         | Nanoindentation<br>Profilometry   | No statistically significant difference in nanoindentation softening between both substrates at the shortest acid-exposure times (2-60 seconds). Profilometry showed that bovine enamel eroding 30% faster than human enamel at longer exposure times (1-60 minutes).  |

Table 3 Summary of *in vitro* studies compared directly between human and bovine teeth in bonding/adhesive strength

| Authors                             | Pertinent aim of the study   | Enamel/<br>Dentin | Type of<br>tooth                      | Adhesive system(s)<br>used   | Root/<br>crown | Type of bond<br>strength tested              | Pertinent outcome   |
|-------------------------------------|--|-------------------|---------------------------------------|--|----------------|--|---|
| Barkmeier and Erickson (72) (1994)  | Evaluate the bond strength of resin composite bonded to both human and bovine teeth.                           | Enamel<br>Dentin  | Permanent                             | Scotch Bond Multi-Purpose  | Crown          | Shear bond strength                          | Bond strengths to bovine enamel were significantly lower than to human enamel.<br>No significant difference between human and bovine dentin bond strength.                                |
| Cadwell and Johannessen (70) (1971) | Compare the adherence of six direct filling materials to human and bovine teeth.                               | Enamel<br>Dentin  | Permanent                             | Did not include the brand names  | Crown          | Ring shear test                              | Adherence of the filling materials to human enamel and dentin significantly higher than that of bovine enamel and dentin.   |
| Fowler et al. (73) (1992)           | Compare both tensile and shear bond strength between human and bovine teeth.                                   | Enamel<br>Dentin  | Permanent                             | Scotchbond 2<br>Ketac-Fil cement   | Crown          | Tensile bond strength<br>Shear bond strength | Bond strength measurements obtained with human and bovine enamel were essentially comparable.<br>A trend for higher bond strength values with bovine than with human dentin was observed. |
| Galhano et al. (81) (2009)          | Compare push out strength of bovine and human teeth.   | Dentin            | Permanent                             | All Bond 2 Universal Adhesive System   | Root           | Push out bond strength                       | Push out bond strength was significantly higher in human root dentin compared to bovine root dentin.  |
| Kaplan et al. (79) (1996)           | Compare bond strength between human and bovine teeth.  | Dentin            | Permanent (human)<br>Primary (bovine) | Scotchbond Bondlite<br>Prisma Universal Bond<br>Gluma  | Crown          | Tensile bond strength                        | Bond strength was lower in bovine dentin than in human dentin.  |
| Krifka et al. (68) (2008)           | Compare the bonding performance of four adhesive luting agents to dentin and enamel of human and bovine teeth. | Enamel<br>Dentin  | Primary                               | Syntac Assortment<br>Adaper Prompt<br>L-Pop<br>iBond Gluma inside<br>Clearfil Protect Bond   | Crown          | Shear bond strength                          | No significant difference in bond strength between human and bovine enamel.<br>No significant difference in bond strength between human and bovine dentin.                                |
| Lopes et al. (74) (2003)            | Compare bond strength between human and bovine substrates using two adhesive systems with different actions.   | Enamel<br>Dentin  | Permanent                             | Scotchbond Multi-Purpose<br>Clearfil Liner Bond 2V   | Crown          | Shear bond strength                          | No significant difference between human and bovine enamel bond strength.<br>Bond strength was significantly higher in bovine dentin when Scotchbond Multi-Purpose was used.               |
| Muench et al. (77) (2000)           | Compare bond strength between human and bovine substrates using three adhesive systems.                        | Dentin            | Permanent                             | Prime and Bond 2.1<br>Single Bond<br>Etch and Prime 3  | Crown          | Tensile bond strength                        | No significant difference between human and bovine dentin bond strength, regardless of the adhesive system used.  |
| Nakamichi et al. (71) (1983)        | Compare adhesive strength to human and bovine teeth using various cements and composite resins.                | Enamel<br>Dentin  | Permanent                             | Carlton<br>Unident<br>HY-Bond Carbo-cement<br>FuyjpieIloIn-oFmer<br>Crown, Bridge &<br>Inlay Cem<br>Adaptic D. Rest<br>Adaptic Bond. A.<br>Clearfil Bond System<br>F | Crown          | Tensile<br>adhesion<br>strength              | No significant difference between human and bovine enamel.<br>No significant difference between human and bovine superficial dentin.  |
| Oesterle et al. (66) (1998)         | Compare bond strength with orthodontic bonding materials between bovine and human teeth.                       | Enamel            | Permanent/<br>primary                 | Orthodontic adhesives (did not include brand name )  | Crown          | shear/peel bond strength                     | Bond strength to primary and permanent bovine enamel was 21% and 44% weaker than permanent human enamel.  |
| Reis et al. (75) (2004)             | Compare bond strength and etched micro-morphology between human and bovine teeth.                              | Enamel<br>Dentin  | Permanent                             | Single Bond Adhesive System  | Crown          | Tensile bond strength<br>SEM                 | No significant difference between human and bovine enamel.<br>No significant difference between human and bovine dentin.<br>Similar dentin morphology for human and bovine teeth.         |
| Retief et al. (78) (1990)           | Compare bond strength between human and bovine teeth.  | Dentin            | Permanent                             | Scotch Bond 2/ Silux   | Crown          | Shear bond strength                          | Shear bond strength significantly greater in human dentin compared to bovine dentin.  |
| Saleh and Tymour (67) (2003)        | Compare bond strength between human and bovine teeth.  | Enamel            | Permanent                             | Reliance Light Bond<br>Fuji Ortho LC   | Crown          | Shear bond strength<br>Tensile bond strength | Shear and tensile bond strength were significantly higher in human teeth compared to bovine teeth.  |
| Saunders (76) (1988)                | Compare the bond strengths of four dentin-bonding agents between human and a bovine substrate.                 | Dentin            | Permanent                             | Scotchbond VLC<br>Topaz<br>Gluma<br>3M experimental agent  | Crown          | Shear impact bond strength                   | No significant difference in bonding strength between human and bovine dentin.  |



Table 3, continuation

| Authors                    | Pertinent aim of the study   | Enamel/<br>Dentin | Type of<br>tooth | Adhesive system(s)<br>used                                     | Root/<br>crown | Type of bond<br>strength tested | Pertinent outcome   |
|----------------------------|--|-------------------|------------------|--|----------------|---------------------------------|---|
| Schilke et al. (80) (1999) | Compare bond strength of dentin adhesives between human and bovine teeth.  | Dentin            | Permanent        | Syntac Bonding system  | Root/<br>Crown | Shear bond strength             | No significant difference between human and bovine coronal dentin. Bonding strength of bovine root dentin was significantly higher than that of human coronal dentin.   |
| Shahabi et al. (65) (1997) | Compare bond strength to enamel of human and bovine teeth.   | Enamel            | Permanent        | Scotchbond MultiPurpose  | Crown          | Shear bond strength             | No significant difference in enamel bond strength between human and bovine enamel.  |
| Titley et al. (69) (2006)  | Compare bond strength over seven and 180 day period of two polyacid modified composite resins bonded to both human and bovine primary teeth. | Enamel<br>Dentin  | Primary          | Dyract AP Etched and Non Etched<br>F2000 Etched and Non Etched | Crown          | Shear bond strength             | No significant difference between human and bovine enamel bond strength was observed irrespective of the storage period. Bond strength of bovine primary dentin was significantly lower than that of human primary dentin, when dentin was etched with 37% H <sub>3</sub> PO <sub>4</sub> . |

Table 4 Summary of *in vitro* studies compared directly between human and bovine teeth in microleakage

| Authors                    | Pertinent aim of the study   | Enamel/<br>Dentin                | Type of<br>tooth | Adhesive system(s)<br>used  | Technique used                | Pertinent outcome  |
|----------------------------|--|----------------------------------|------------------|---|-------------------------------|--|
| Abuabara et al. (7) (2004) | Evaluate the marginal microleakage of human and bovine teeth restored with composite and glass inomer.                   | Enamel margins                   | Permanent        | Single Bond   | Spectrophotometer             | Microleakage was significantly higher in bovine teeth compared to human teeth.                     |
| Almeida et al. (85) (2009) | Analyze the influence of human and bovine substrates on marginal microleakage.   | Enamel margins                   | Permanent        | Prime & Bond 2.1 Adhese   | Stereomicroscope              | No significant difference between human and bovine substrates.                                     |
| Camargo et al. (86) (2007) | Compare the pulp chamber penetration of hydrogen peroxide between humane and bovine teeth.                               | Pulp chamber wall                | Permanent        | Composite resin<br>Glass ionomer cement<br>Resin modified glass ionomer | Spectrophotometer             | Significant higher peroxide penetration into pulp chamber in human teeth compared to bovine teeth. |
| Fitchie et al. (84) (1995) | Compare the microleakage of bovine and human teeth using one adhesive system and two different types of composite resin. | Enamel margins<br>Dentin margins | Permanent        | Syntac  | <sup>45</sup> Ca radioisotope | No significant difference between human and bovine substrates.                                     |
| Lopes et al. (82) (2009)   | Compare the microleakage pattern between bovine and human teeth.   | Dentin margins                   | Permanent        | Clearfil SE Bond<br>Scotchbond 1  | Confocal microscopy           | Bovine teeth showed significantly higher percentage of leakage compared to human teeth.            |
| Reeves et al. (83) (1995)  | Compare the microleakage behavior of 3 bonding systems in human and bovine teeth.  | Enamel margins<br>Dentin margins | Permanent        | Scotchbond Multi-Purpose<br>All Bond 2<br>Universal Bond 3              | <sup>45</sup> Ca radioisotope | No significant difference between human and bovine substrates.                                     |
| Retief et al. (78) (1990)  | Evaluate the quantitative microleakage of human versus bovine substrates.  | Dentin margins                   | Permanent        | Scotchbond 2  | Spectrophotometer             | Microleakage was significantly greater in bovine teeth than in human teeth.                        |

in dentin of human and variously aged bovine teeth. However, Knoop hardness of human enamel was similar to bovine enamel in older age groups (38 and 48 months), but significantly higher than that of younger age groups (20 and 30 months).

### Dental caries studies

Table 1 shows the details of the seven studies reviewed (6,9,50-54). There were four *in vitro* studies (6,9,50,51), one *in situ* study (52), and two *in vitro/in situ* studies (53,54). Only one study in this category compared dentin caries between human and bovine teeth (52), whereas the

remaining studies compared only enamel caries between human and bovine teeth (6,9,50-54). Inconsistent data was found regarding the use of bovine substrates as an alternative to human teeth in dental caries studies.

### Dental erosion/abrasion studies

Table 2 illustrates the 10 studies reviewed in this category (55-64). There were three *in situ* studies (55-57), and seven *in vitro* studies (58-64). Two *in situ* studies compared between human and bovine enamel (55,56); the other one compared enamel and dentin of human and bovine teeth (57). Four *in vitro* studies compared human

and bovine enamel (58-61), whereas, the remaining three *in vitro* studies compared between human and bovine dentin (62-64). Inconsistent outcomes were suggested regarding the use of bovine substrate as an alternative to human substrate in dental erosion/abrasion studies.

### Bonding/adhesive strength studies

Table 3 depicts the 17 *in vitro* studies reviewed in this category (65-81). Three studies tested only human and bovine enamel (65-67). Eight studies compared both enamel and dentin of human and bovine teeth (68-75). The remaining six studies compared only dentin bond strength between human and bovine teeth (76-81). Inconsistent recommendations were proposed regarding the use of bovine substrate as an alternative to human substrate in bonding/adhesive strength studies.

### Microleakage studies

Table 4 illustrates seven *in vitro* studies reviewed in this category (7,78,82-86). Three of the seven studies found that microleakage values were higher in bovine compared to human substrates, both in enamel (7) and dentin (78,82). On the other hand, three studies found no significant differences in marginal microleakage between human and bovine teeth, both in enamel (83-85) and dentin (83,84). Camargo et al. (86) showed that the penetration of 38% hydrogen peroxide bleaching agent into pulp chambers was higher in human teeth compared to bovine teeth, regardless of the restorative material used.

Based on the findings of 68 selected articles in this review, inconsistent data exist regarding whether bovine teeth can be considered appropriate substitute for human teeth, in the reviewed fields of dental research. Morphological, chemical composition and physical property differences between human and bovine teeth must be considered when interpreting results obtained from any experiment with bovine teeth substrate.

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