0099-2399/88/1401-0035/$02.00/0
JOURNAL OF ENDODONTICS

Printed in U.S.A.
VOL. 14, NO. 1, JANUARY 1988

CLINICAL ARTICLE

Darkfield Microscopy as a Diagnostic Aid in Differentiating Exudates from Endodontic and Periodontal Abscesses

Martin Trope, DMD, Leif Tronstad, DMD, PhD, Edward S. Rosenberg, DMD, and Max Listgarten, DDS

It was the purpose of this study to determine if a periodontal abscess could be differentiated from an endodontic abscess by the types and proportion of microorganisms found in the abscess exudate in darkfield microscopic examination. Dental abscesses with draining sinus tracts in 17 patients were studied. Eight of the abscesses were diagnosed clinically to be of endodontic origin and nine of periodontal origin. When the occurrence of motile rods in endodontic and periodontal abscesses were compared, the differences were not significantly different (mean, 12.1 ± 10.8% and 7.5 ± 3.9%, respectively). In abscesses diagnosed clinically as periodontal, spirochaetes were the predominant cell (mean, 40.6 ± 10.9%) with coccoid cells present in significantly lower numbers (mean, 19.7 ± 10.9%). In endodontic abscesses the reverse was true. Coccoid cells dominated (mean, 44.3 ± 19.7%) and only few spirochetes were present (mean, 5.6 ± 4.7%). The range of percentages for coccoid cells was 7 to 40% in periodontal abscesses and 24 to 84% in endodontic abscesses. The overlap of range of 25 to 40% limited the use of these organisms for diagnostic purposes. For the spirochetes, on the other hand, there was a distinct difference in the range of percentages in the two types of abscesses. In periodontal abscesses the occurrence of spirochetes ranged from 30 to 60%, whereas in endodontic abscesses the range was 0 to 10%. Thus, the percentage of spirochetes as seen by darkfield microscopy may be of value in the differential diagnosis of periodontal and endodontic abscesses.

The differentiation between endodontic and periodontal abscesses is sometimes difficult. This is especially true when the vitality of the tooth in question cannot be used as an aid in the diagnosis because of previous endodontic treatment or a full crown restoration, and when the abscess in question drains through the gingival sulcus.

Darkfield microscopic analysis of the proportions of different bacteria within a periodontal pocket has been used in periodontics as a chairside diagnostic and monitoring method to ascertain the status of periodontal lesions (1, 2). It is a quick, simple, and relatively inexpensive technique of classifying bacterial samples according to bacterial shape, size, and motility (1). It has been found that sites with periodontal disease have significantly higher proportions of spirochetes and motile rods and lower proportions of cocci than periodontally healthy sites (1–3). Furthermore, it has been shown that these proportions will be reversed following adequate periodontal therapy (2).

Thus, the presence of spirochetes is a characteristic trait of sites of adult periodontitis, with percentages between 29 and 57% of the total bacterial flora having been reported (4–7). On the other hand, spirochetes have not been found (8–10) or have been found in only low percentages (11) in the microflora of infected root canals.

It was the purpose of the present study to compare the microflora of periodontal and endodontic abscesses to establish if differences exist that could be used for differential diagnostic purposes.

MATERIALS AND METHODS

Dental abscesses with draining sinus tracts in 17 patients between 32 and 57 yr of age were studied. Eight of the abscesses were diagnosed clinically to be of endodontic origin and nine were judged to be periodontal abscesses. When the sinus tract drained through the alveolar mucosa, exudate was milked onto a clean periodontal currette or spoon excavator. In the cases of drainage through the sulcus, exudate was

collected according to the method described by Listgarten and Hellden (1). A sterile periodontal currette or spoon excavator was introduced through the sulcus or pocket orifice as far apically as possible and the bacterial contents were removed. If necessary in both situations, the process was repeated several times to obtain enough material. The samples were suspended in 0.3 ml of sterile 0.85% sodium chloride solution containing 1% gelatin by vigorously agitating the tip of the instrument in the solution. The bacterial suspensions were dispersed just prior to the examination by aspirating and expelling the fluid three times through a disposable tuberculin syringe attached to a 23-gauge needle. One drop of the suspension was then applied to a microscopic slide and coverslipped. Excess fluid was removed by inverting the slide over an absorbent surface and applying moderate pressure.

The slide was then examined by darkfield microscopy at a magnification of ×1200. If the preparation was too dense, the sample was further diluted with saline until about 100 to 200 bacteria were present in the specimen examined. The bacteria were classified according to the method of Listgarten and Hellden (1) as follows: *Coccoid cells:* Cells were approximately 0.5 to 1.0 μm in diameter showing a bright outline with a dark center. Also included in this group were coccobacillary forms, the length of which was up to, but not more than twice the width of the cell (Fig. 1). *Spirochetes:* Three varieties of these cells were counted. *Small spirochetes:* Helicoidal cells were approximately 0.2 to 0.3 μm in width and up to 10 μm in length with a single-contoured outline and relatively tight coils (Fig. 2). *Intermediate spirochetes:* Helicoidal cells were approximately 0.3 to 0.4 μm in width and up to 15 μm in length. The cell outline was single-contoured by darkfield and the coiling not as tight as for small spirochetes (Fig. 3). *Large spirochetes*: Helicoidal cells were 0.5-μm wide or wider and up to 20 μm in length. The cells had a wavy rather than a coiled appearance (Fig. 4). *Motile rods:* This category included all cell types, other than spirochetes which exhibited motility by darkfield. A flagellated curved rod is shown in Fig. 5. Cell motility was distinguished from Brownian motion and the streaming of fluid between the slide and the coverslip, by its purposeful pattern. *Straight rods*, filaments, fusiforms, and curved rods were grouped together and made up the rest of the count.

The groups were statistically compared using a *t* test for independent samples. A level of confidence for significance of $p < 0.05$ was used.

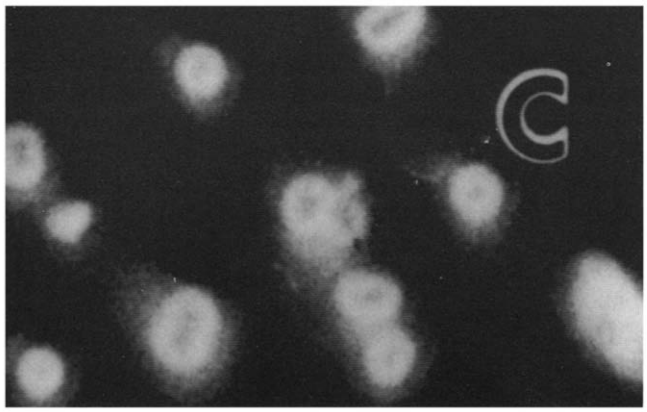

FIG 1. Darkfield microscopic appearance of coccoid (*C*) cells. The cells are 0.5 to 1.0 μm in diameter with a bright outline and dark center (original magnification ×1,200).

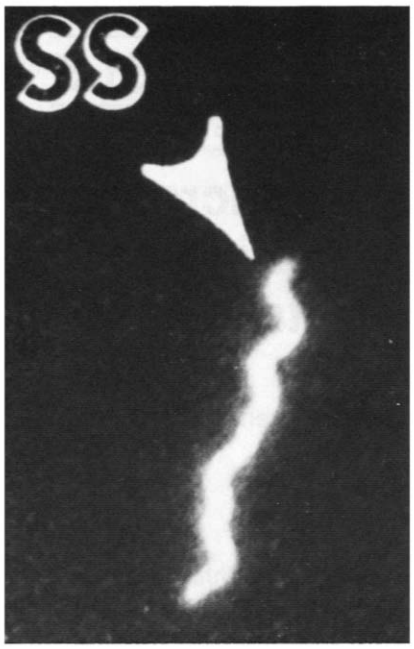

FIG 2. Darkfield microscopic appearance of small spirochete (*SS*). The cells are helicoidal, approximately 0.2 to 0.3 μm in width and up to 10 μm in length with a single-contoured outline and relatively tight coils (original magnification ×1,200).

FIG 3. Darkfield microscopic appearance of intermediate spirochete. These cells are helicoidal, approximately 0.3 to 0.4 μm in width and up to 15 μm in length. The cell outline is single-contoured with coiling not as tight as for small spirochetes (original magnification ×1,200).

RESULTS

The percentage of coccoid cells, spirochetes, and motile rods in abscesses diagnosed to be of periodontal origin are presented in Table 1. The spirochetes were the predominant cell type, with a mean proportion of 40.6 ± 10.9%. The coccoid cells (mean proportion, 19.7 ± 10.9%) were found in significantly lower proportions than the spirochetes. The mean proportion of motile rods was 7.5 ± 3.9%.

The percentages of bacteria found when the abscesses were diagnosed to be of endodontic origin are presented in Table 2. In this type of abscess the predominant organisms were the coccoid cells, with a mean of 44.3 ± 19.7%. Motile rods were found in significantly lower proportions (mean, 12.1 ± 10.8%). Spirochetes made up only a small proportion of the bacteria in endodontic abscesses (mean, 5.6 ± 4.7%).

For purposes of comparison, the percentages of coccoid cells, spirochetes, and motile rods in periodontal and endodontic abscesses are shown together in Table 3. When the occurrences of motile rods were compared, there was no significant difference between

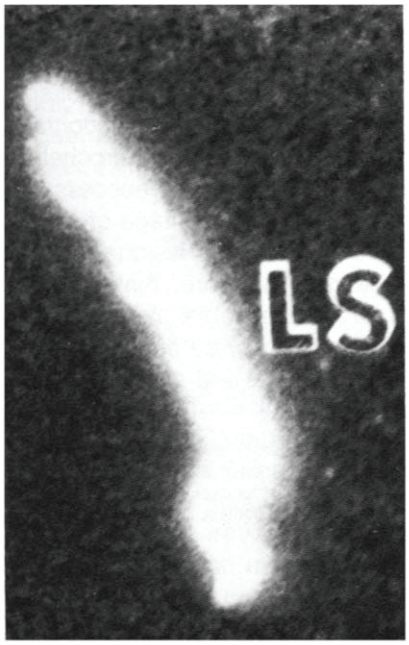

FIG 4. Darkfield microscopic appearance of large spirochete (*LS*). The cells are helicoidal, 0.5-μm wide or wider and up to 20 μm in length. The cells have a wavy rather than a coiled appearance (original magnification ×1,200).

FIG 5. Darkfield microscopic appearance of curved rod. This cell is classified as a motile rod because of the presence of a flagellum (original magnification ×1,200).

TABLE 1. Proportions (percent) of bacteria in periodontal abscesses as determined in darkfield microscopy

	Coccoid Cells	Spirochetes	Motile Rods
	18	58	10
	12	45	4
	15	45	3
	40	30	10
	19	30	5
	32	30	10
	7	38	14
	9	56	9
	26	34	3
Mean	19.7	40.6	7.5
SD	±10.9	±10.9	±3.9

TABLE 2. Proportions (percent) of bacteria in endodontic abscesses as seen by darkfield microscopy

	Coccoid Cells	Spirochetes	Motile Rods
	28	10	6
	25	9	29
	35	10	8
	84	8	0
	60	0	0
	32	8	24
	50	0	19
	41	0	11
Mean	44.3	5.6	12.1
SD	±19.7	±4.7	±10.8

TABLE 3. Comparison of proportions (percent) of bacteria in abscesses of periodontal and endodontic origin as seen by darkfield microscopy

	Coccoid Cells		Spirochetes		Motile Rods	
	Perio*	Endo	Perio	Endo	Perio	Endo
	18	28	58	10	10	6
	12	25	45	9	4	29
	15	35	45	10	3	8
	40	84	30	8	10	0
	19	60	30	0	5	0
	32	32	30	8	10	24
	7	50	38	0	14	19
	9	41	56	0	9	11
	26		34		3	
Mean	19.7	44.3	40.6	5.6	7.5	12.1
SD	±10.9	±19.7	±10.9	±4.7	±3.9	±10.8

* Perio, periodontal; endo, endodontic.

TABLE 4. Range of bacterial (percentages) in abscesses of periodontal and endodontic origin as seen by darkfield microscopy

	Periodontal Abscesses	Endodontic Abscesses
Coccoid cells	7–40	25–84
Spirochetes	30–58	0–10*

* Significantly different, $p < 0.05$.

the two types of abscesses (7.5 ± 3.9% in periodontal abscesses versus 12.1 ± 10.8% in endodontic lesions). Significantly more coccoid cells were found in the endodontic (44.3 ± 19.7%) than in periodontal lesions (19.7 ± 10.9%).

Spirochetes, on the other hand, were significantly more abundant in periodontal (40.6 ± 10.9%) than in endodontic abscesses (5.6 ± 4.7%). The range of percentages of coccoid cells and spirochetes in the two types of abscesses is presented in Table 4. The coccoid cells contributed between 7 and 40% of the flora in the periodontal abscesses and 25 and 84% of the flora of the endodontic abscesses. The ranges of spirochetes were 30 to 58% in periodontal lesions and 0 to 10% in endodontic lesions.

DISCUSSION

The technique of darkfield analysis can easily be learned by the dentist or auxiliary personnel and is simple to carry out. An advantage of the procedure is that the results can be obtained chairside in a few minutes, so that definitive treatment can begin at the initial visit which often is associated with excruciating pain.

For the purposes of this study only abscesses where a correct diagnosis could confidently be made were used. Because of this the proportions of the various types of bacteria as they occurred in the periodontal and endodontic abscesses could be assessed and meaningful comparisons made.

For the periodontal abscesses, the range of spirochetes was between 30 and 58%. This range of percentages was similar to the range found when asymptomatic but active sites of adult periodontitis were studied (4–7). For the endodontic abscesses, the range of spirochetes was between 0 and 10%. Here also the range of percentages corresponded well with the percentages of spirochetes in infected root canals (8–10). It is interesting to note that the route of drainage had little bearing on the results. In eight of nine periodontal abscesses, the exudate drained through the sulcus and in one instance through a sinus tract in the alveolar mucosa. In this one case the spirochete count at 34% was similar to the counts in the other periodontal abscesses. Three of the eight endodontic abscesses drained through the gingival sulcus. In these lesions the spirochetes count was between 8 and 10% and accordingly within the range of the other endodontic abscesses where drainage occurred through the alveolar mucosa.

Significant differences in the percentages of both coccoid cells and spirochetes were found between the two types of abscesses. To be valuable as a quick diagnostic method, the range of percentages of the various bacteria in the different abscesses must not overlap. With regard to coccoid cells there was an overlap in the range of 25 to 40%. Thus, a lesion could have been both periodontal or endodontic in origin. In fact, three of the periodontal and four of the endodontic lesions fell within this range. Thus, the use of the percentages of cocci for differential diagnostic purposes appeared to be of limited value.

On the other hand, the proportions of spirochetes in the floras of endodontic and periodontal abscesses were markedly different. If the spirochete count was around 10% or lower, then the abscess was of endodontic origin and if the percentage of spirochetes was around 30% or above, the lesion was a periodontal abscess. It seems, therefore, that the percentage of spirochetes of the microbial flora as determined in darkfield microscopy may be of value in the differential diagnosis of periodontal and endodontic abscesses.

Dr. Trope is assistant professor and director of doctoral endodontics, Dr. Tronstad is professor and chairman of endodontics, Dr. Rosenberg is associate professor and director of postdoctoral periodontics, and Dr. Listgarten is professor and chairman of periodontics, School of Dental Medicine, University of Pennsylvania, Philadelphia, PA.

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