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THE RELATIVE PROPORTIONS OF SODIUM, POTASSIUM AND CALCIUM IN GINGIVAL POCKET FLUID

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A flow of tissue fluid through the epithelium in clinically healthy gingival pockets has been demonstrated in dogs by *Brill & Krasse* (1958) and in man by *Brill & Björn* (1959). This flow of tissue fluid may be due either to a simple filtration of tissue fluid through epithelial layers or it may be an exudation from underlying inflammation.

Even in the clinically healthy gingivae an infiltration of round cells often occurs in the sub-epithelial tissue, and this is taken to indicate the presence of chronic inflammation. In extra-cellular fluid in damaged tissues, reduced sodium and increased potassium concentrations occur because of a shift of sodium into the cells and a shift of potassium out into the extra-cellular fluid (*Letterer* 1959, *Fuhrman* 1960). With this observation in mind, it is of interest to study the relationship between Na and K in gingival pocket fluid. Various experiments have shown that the blood plasma is the original source of the fluid (*Brill & Krasse* 1958, *Brill & Björn* 1959, *Brill* 1959 a). If the fluid passes through undamaged connective tissue and epithelium, it should contain the same proportions of sodium and potassium as plasma and interstitial fluid (extra-cellular fluid). On the other hand, if it passes through damaged tissue, decreased sodium/potassium ratio should be found according to the observations cited above. Thus, the relation between sodium and potassium ought to indicate whether the pocket fluid from clinically healthy gingivae

is to be regarded as a simple filtration product or rather as an inflammatory exudate. For comparison exudate from chronically inflamed gingivae may be studied.

MATERIAL AND METHODS

Two groups of persons were studied. The first group consisted of 17 dental nurses ranging in age from 18 to 23 years with no clinical evidence of gingivitis. The second group was composed of patients of the Dental School and contained 11 males and 13 females between 17 and 74 years of age. All of the patients had chronically inflamed gingivae, with or without alveolar bone loss.

The fluid samples were collected from the gingival pockets of upper premolars and first molars in the dental nurse group and from upper incisors, canines and premolars in the patient group. The reason why premolars and molars were used in the dental nurse group was that it proved difficult to obtain sufficient amounts of fluid from the gingival pockets of incisors and canines in this group. The operation field was carefully dried with cotton rolls and compressed air. In order to get sufficient amounts of fluid, this was allowed to accumulate in the pocket for about 15 minutes. It was then collected by means of a capillary tube, which was placed in the opening of the gingival pocket, care being taken to avoid damaging the gingivae. The tube was gently moved back and forth and gentle suction through a rubber tube was applied.

In the dental nurse group, samples were first collected with the gingivae in a resting state. For comparison samples were afterwards collected after mechanical stimulation of the gingivae with a blunt instrument, it being known that brief mechanical stimulation increases the amount of gingival fluid (*Brill & Krasse*, 1959). After stimulation, adequate samples could be taken immediately. In the group with chronic inflammation, the samples were taken without stimulation. In most cases sufficient amounts of fluid could be collected immediately after preparation of the operation field.

The fluid was blown out from the capillary tube into 1.5 ml of redistilled water in a centrifuge tube.

In order to remove suspended cells the samples were centrifuged before the analysis. Control experiments, however, showed

that this procedure did not influence the results and the centrifuging was therefore later abandoned. Instead, the tubes were allowed to stand for 10 minutes before the analysis was carried out.

Analysis of Na, K and Ca was performed by flame photometry in an Eppendorf flame photometer. Because of the difficulties in measuring the small amounts of fluid obtained from the pockets, only the relative proportions of the ions could be determined. The ratios are calculated from values in m.Eq/litre. The sodium concentration in the dilutions analysed ranged from 8—220 m.Eq/litre; the potassium and calcium from 0.25—10 and 0.2—1.5 m.Eq/litre respectively.

RESULTS

The relative proportions of Na/K, Na/Ca and K/Ca in gingival pocket fluid before and after mechanical stimulation of the gingivae are shown in Table 1.

Table 1

Relative proportions of sodium, potassium and calcium in gingival pocket fluid from clinically healthy gingivae before and after mechanical stimulation.

The ratios calculated from values in m.Eq./litre

Subject	Before stimulation			After stimulation		
	Na/K	Na/Ca	K/Ca	Na/K	Na/Ca	K/Ca
1	6.4	18.2	2.8	6.3	19.1	3.0
2	5.2	13.8	2.6	6.3	21.8	3.5
3	5.3	10.0	1.9	4.2	21.4	5.1
4	3.5	16.2	4.6	2.6	16.7	6.3
5	2.6	5.2	2.0	0.8	5.7	7.3
6	2.3	7.6	3.2	3.5	12.4	3.5
7	4.8	6.8	1.4	2.8	—	—
8	2.6	8.4	3.3	4.7	20.9	4.4
9	3.3	8.0	2.4	4.5	17.0	3.7
10	8.8	15.0	1.7	8.2	19.8	2.4
11	0.6	7.1	12.6	1.1	8.8	7.7
12	2.8	9.7	3.4	2.9	16.4	5.6
13	1.2	9.1	7.4	2.9	13.5	4.7
14	9.0	23.2	2.6	5.0	23.3	4.6
15	1.7	5.5	3.2	3.0	12.7	4.2
16	3.9	5.7	1.5	4.7	3.0	0.6
17	2.4	—	—	2.3	—	—
Mean	3.9	10.6	3.5	3.9	15.5	4.4
Standard deviation	± 2.4	± 5.2	± 2.8	± 1.9	± 6.1	± 1.8

The values from chronically inflamed gingivae are given in Table 2.

Table 2

Relative proportions of sodium, potassium and calcium in gingival pocket fluid from chronically inflamed gingivae.

Subject	Na/K	Na/Ca	K/Ca
1	8.4		
2	1.3		
3	2.7		
4	10.3		
5	9.5		
6	2.6		
7	8.7	25.6	2.9
8	1.7	5.1	3.0
9	5.1	7.3	1.4
10	18.3		
11	5.8	11.5	2.0
12	17.4		
13	9.7		
14	6.3	15.6	2.5
15	22.9	35.3	1.5
16	5.2	17.5	3.3
17	29.1	33.7	1.2
18	10.6	26.2	2.5
19	7.6		
20	10.2		
21	9.7	11.4	1.2
22	12.9		
23	7.4		
24	12.1	12.4	1.0
Mean	9.8	18.3	2.1
Standard deviation	± 6.6	± 10.4	± 0.8

Table 3

Relative proportions of sodium, potassium and calcium in gingival pocket fluid, extra-cellular fluid and intra-cellular fluid**.*

	Na/K	Na/Ca	K/Ca
Clinically healthy gingiva, before stimulation	4/1	11/1	4/1
Clinically healthy gingiva, after stimulation	4/1	16/1	4/1
Chronically inflamed gingiva	10/1	18/1	2/1
Extra-cellular fluid plasma, interstitial fluid)	28/1	28/1	1/1
Intra-cellular fluid	1/50	0.8/1	40/1

* The values used have been calculated from Gamble (1947)

** The values used have been calculated from values given by Conway (1957)

A comparison between the rounded mean values from Tables 1 and 2 is shown in Table 3. In this table, the relative proportions of sodium, potassium and calcium in extra-cellular and in intra-cellular fluid have been included.

From Tables 1 and 3 it is clearly seen that the relative proportions of Na/K are practically identical in samples from clinically healthy pockets before and after stimulation. A comparison between these values and the corresponding ratio for extra-cellular fluid shows that the Na/K ratio is considerably lower in gingival pocket fluid. This indicates a higher concentration of potassium and/or a lower concentration of sodium. The ratio is, however, still far above that found in intra-cellular fluid.

From Tables 2 and 3 it can be seen that the Na/K ratio in fluid from chronically inflamed gingivae is more than two times that in fluid from clinically healthy gingivae, but still much lower than the ratio in extra-cellular fluid. Because of the small amounts of suspension used for analysis, calcium could not be determined in all cases. In addition, in some cases the calcium concentration was at the lower limit of the capacity of the flame photometer and thus the ratios Na/Ca and K/Ca are not fully reliable. In these quotients, however, the same tendency is found as with the Na/K ratio, i.e. that the values for the chronically inflamed gingivae when compared with those from the clinically healthy gingivae tend towards the extra-cellular fluid ratios.

DISCUSSION

In gingival pocket fluid from both clinically healthy and chronically inflamed gingivae the Na/K ratio was considerably lower than is found in extra-cellular fluid. Since the blood plasma is the original source of the fluid, the difference in Na/K ratio between plasma and gingival pocket fluid is of interest. The high proportion of potassium in gingival pocket fluid could depend on some error in experimental technique and this question must therefore be considered first.

Great numbers of leukocytes and epithelial cells can occur in the gingival pocket fluid (*Sharry & Krasse,* 1960) and, when the

flame photometry is performed, these cells could be burnt in the flame and thus give rise to the higher proportion of potassium. Against this explanation the following facts must be weighed: —

1. In control experiments with fairly heavy suspensions of blood leukocytes or salivary sediment the Na/K ratios did not differ when the cells were left suspended or when they were removed by centrifugation.
2. Centrifuging the test samples did not change the Na/K ratio.
3. Fluid from stimulated pockets contains far fewer cells than that from unstimulated pockets (*Egelberg,* 1962), but the Na/K ratio is practically the same.

It can thus be assumed that the increased proportion of potassium does not depend on burning of cells.

Another explanation is that intra-cellular potassium has been released from the cells when they were suspended in re-distilled water. However, this would have given a higher proportion of potassium in fluid from unstimulated pockets where the number of cells is much greater than in the stimulated ones. This did not prove to be the case.

Thus, it is most probable that a high proportion of potassium really occurs in gingival pocket fluid. This shows that intracellular potassium is added to and/or sodium withdrawn from the extra-cellular fluid on its way out into the gingival pocket. As an increased potassium concentration is found in exudate *Menkin* 1956) and in extra-cellular fluid of damaged tissues (*Fuhrman* 1960) these observations suggest that the gingival pocket fluid, even from clinically healthy gingivae is derived from tissues with altered metabolism. This would mean that the gingival pocket fluid cannot be regarded as a simple filtration product but rather as an inflammatory exudate. This conclusion is supported by the frequent presence of round cell infiltration in subepithelial tissues in clinically healthy gingivae.

It should be mentioned that a sodium potassium shift may also occur when the fluid passes the degenerating and shedding cells which are found in superficial layers of pocket epithelium (*Waerhaug* 1952, *McHugh* 1961). These cells could thus act as

a membrane which influences the composition of the fluid as it passes through. The relative importance of degenerating epithelial cells for the Na/K ratio can be discussed on the basis of the results obtained from stimulated and unstimulated gingivae. In both series the Na/K and K/Ca ratios were practically the same. Stimulated pockets produced a larger amount of fluid per unit of time and the potassium ratio ought to have been decreased if the sodium potassium shift depended on the degenerating superficial cells. Thus, the fluid from stimulated pockets does not differ from that of unstimulated pockets with regard to Na/K ratio in spite of its increased volume. This would mean that stimulation alters the sodium potassium shift as well as it increases the exudation.

The Na/K ratio in fluid from chronically inflamed gingivae was somewhat higher than that from clinically healthy gingivae but still lower than in extra-cellular fluid. The Na/Ca and K/Ca values also tend towards the extra-cellular fluid ratios (see Table 3). This might be due to the increased vascularisation and ulceration of the pocket epithelium which occur in inflamed gingivae. These changes lead to an increased exudation, probably from a great number of superficial blood vessels with increased permeability. Hence, the fluid does not pass the same amount of tissue as in healthy gingivae and thus the better agreement with the plasma values can easily be understood. This theory is supported by the observation that two persons having an extremely large flow of gingival pocket fluid showed very high Na/K and Na/Ca ratios and low K/Ca ratio (Table 2, subjects 15 and 17).

It is difficult to say whether or not the increased proportion of potassium is of any importance for the host-microbial relationship in the gingival pocket. However, it is of great interest to note that *Pfeffer* (1904) claimed that potassium salts could produce positive tactic responses in spirilla, since it is well known that vibrio-like organisms and spirochetes occur in great numbers especially in the gingival pocket. With regard to the antimicrobial effect of the gingival pocket fluid the following facts must be considered. A great number of polymorphonuclear leukocytes are regularly found in the gingival pocket (*Sharry & Krasse* 1960). The pocket fluid contains different serum protein

components including gammaglobulins (*Ewl & Brönnestam*, 1960). By the simultaneous presence of leukocytes and gammaglobulins (opsonins) it is possible that phagocytosis could occur in the gingival pocket. If this is the case, the presence of Ca^{++}-ions could be of importance, since it has been shown that motility and peptidase activity of the polymorphonuclear leukocytes is increased by this ion (*Suter* 1956). Whether the changed Na/K ratio has any influence on the potential phagocytotic activity no information could be obtained from the literature. The dynamic host-microbial relationship in the gingival pocket is probably determined by a series of factors, one of which might be the ion relations. To throw light on this problem further investigations are necessary.

SUMMARY

The relative proportions of sodium, potassium and calcium were studied in gingival pocket fluid.

In comparison with the original source of the fluid, the blood plasma, a proportional increase of potassium was found in the pocket fluid from clinically healthy gingivae. This observation indicates that intra-cellular potassium is added to the extra-cellular fluid on its way out into the gingival pocket. As an increased potassium concentration is found in extra-cellular fluid of damaged tissues these observations suggest that gingival pocket fluid, even from clinically healthy gingivae, is derived from tissues with altered metabolism. This would mean that gingival pocket fluid cannot be regarded as a simple filtration product but rather as an inflammatory exudate.

The implications of the findings for the host-microbial relationship are discussed.

RÉSUMÉ

PROPORTIONS RELATIVES DE SODIUM, POTASSIUM ET CALCIUM DANS LE LIQUIDE DU CUL-DE-SAC GINCIVAL.

Les proportions relatives de sodium, de potassium et de calcium ont été étudiées dans le liquide des culs-de-sac gingivaux.

Par comparaison avec la source originelle du liquide, le plasma sanguin, une augmentation des proportions de potassium a été

trouvée dans le liquide des culs-de-sac de gencives cliniquement saines. Cette observation indique que le potassium intra-cellulaire s'ajoute au liquide extra-cellulaire pendant sa sortie dans le cul-de-sac gingival. Etant donné qu'une augmentation de la concentration du potassium s'observe dans le liquide extra-cellulaire de tissus lésés, ces observations laissent à supposer que le liquide du cul-de-sac gingival, même en provenance de gencives cliniquement saines, provient de tissus ayant un métabolisme altéré. Cela indiquerait que le liquide du cul-de-sac gingival ne peut être considéré comme le produit d'une simple filtration, mais plutôt comme un exsudat inflammatoire.

Les conséquences de ces résultats en ce qui concerne les relations entre l'organisme et les hôtes microbiens font l'objet d'une discussion.

ZUSAMMENFASSUNG

DIE RELATIVEN PROPORTIONEN DES NATRIUMS, KALIUMS UND KALZIUMS IN DER GEWEBEFLÜSSIGKEIT DER ZAHNFLEISCHTASCHE

Im Vergleich mit dem Blutplasma, aus welchem ursprünglich die Flüssigkeit herstammt, zeigte die Gewebeflüssigkeit der Zahnfleischtasche vom klinischen gesunden Zahnfleisch eine Zunahme des Kaliums.

Diese Observation deutet darauf, dass intracelluläres Kalium zu der extracellulären Flüssigkeit auf dem Wege zur Zahnfleischtasche hinzugefügt wird.

Da eine erhöhte Kaliumkonzentration in der extracellulären Flüssigkeit des schadhaften Gewebes gefunden ist, deuten diese Observationen darauf, dass die Gewebeflüssigkeit der Zahnfleischtasche auch vom klinisch gesunden Zahnfleisch von einem Gewebe mit verändertem Metabolismus kommt.

Dieses zeigt, dass die Gingivalflüssigkeit nicht als ein Filtrationsprodukt sondern vielmehr als ein inflammatorisches Exsudat betrachtet werden kann.

Die Bedeutung dieser Observationen für das Verhältnis zwischen dem Mikrobenträger und der Mikrobe wurden diskutiert.

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