INFLUENCE OF SOME SOLUTIONS ON THE RATE OF PERMEABILITY IN ZEA MAYS SEEDS

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(with 2 Text-Figures.)

INDEX.

I. Introduction.

Every organism, whether plant or animal, receives from its surroundings substances in some form or another. These substances are for the most part in the form of low concentrated solutions. Many workers have shown that permeability of water and solutions depend, besides other factors, on the type of ions present in the media. It would be interesting to observe the rate of permeability of water out of various solution or solutions as they are taken in by an organism. It is probably true that in many problems of permeability isolated cells, tissues and even organs have been employed. However, it seems to the writer that the information gained from such studies may not be applicable to an organism as a whole, since all cells and tissues possessed by higher plants may not be alike in structure and function. It must be pointed out that the employment of isolated cells or tissues is preferable, because it allows the use of more rigid control and precise methods of attack, which higher organisms, as a whole, do not permit.

Since seeds come near an ideal and satisfactory material for the quantitative study of permeability from the point of view of exact control and method (although not like isolated cells), they have attracted the attention of plant physiologists. In experiments reported in this paper, seeds have been employed with the hope that they might yield some data, which would add to the innumerable contributions already made in this perplexing phase of plant physiology. Barton-Wright, on this question, writes thus, "The whole question of the permeability of plant cells in general is in a very unsatisfactory position."

II. Review of Literature.

Two kinds of walls have been recognized in relation to permeability. One of them is almost non-permeable to water. Priestley (1921) and Collins (1918) attribute this characteristic to its cutinized or suberized layers. The other wall is permeable to water but to a greater degree impermeable to solutes such as sodium chloride. Gola (1905), Brown (1907), Shull (1913), Tjebbes (1912), Rippel (1918), Gassner (1915), Schroeder (1911) and many other workers have experimentally suggested the presence of semi-permeable layer in many species of seeds.

Brown (1909) has shown that barley seeds allow less absorption of water from salt solutions than from pure water. Rippel (1918) seemed to have found a direct relationship between the intake of table salt and water.

There are certain authors, whose findings in respect to water absorption do not seem to agree with Brown's (1909) views. For instance, Thoday (1918) has found an increase in the rate of water absorption at lower concentrations of the solute as compared with that in distilled water. He used osmic acid and mercuric chloride.

How one would account for the conflicting results reported by Thoday (1918), Brown (1909) and Rippel (1918)? It seems to the writer that the walls enclosing seeds may have different characteristics not only due to difference in species but also due to their variable stages of development within the species. Such variations may allow unequal rate of permeability. Stiles (1924) in reviewing this subject holds a somewhat similar view since he remarks on permeability with reference to the properties of the cell walls as thus, "It is clear that we may find walls with very different degrees of permeability both to water and to various dissolved substances."

Stiles and Jorgensen (1917) studied the permeability of N/8 and N/3 sodium chloride and various concentrations of ethyl alcohol in potato tissue. They found that with sodium chloride the tissue neither gained nor lost in weight. In M alcohol the maximum intake of water reached in about 7 or 8 hours, while in 2M alcohol the maximum point reached in about half an hour. Thoday obtained similar curves for chloroform and mercuric cyanide as Stiles and Jorgensen did for alcohol.

Brown and Worley (1912) have noted the entrance of ethyl acetate in barley seeds. Rapid rate of water absorption was observed in these grains where ethyl acetate solution was employed; while there was a slower intake of water where pure water alone was used. Similar tendency has been recorded by Brown and Tinker (1916). Such a variation in the rate of water permeability, may be due to the differences of the surface tensions of the external media, as suggested by Brown and Worley.

Scarth (1925) observed the entrance of ions in Spirogyra cells. He found that many di- and trivalent cations are taken in rapidly at first, afterwards the rate slows down to a standstill. According to him, the penetration of an ion is determined by two opposing forces within the cell, namely, the one tending to the entrance of the ion and the other forcing its exclusion.

Permeability of ions varies as has been noted by Redfern (1922), who found that the penetration of calcium and chlorine ions is different in the case of pea seedlings. Similarly Haas and Reed (1926) showed recently that two ions were not absorbed in equivalent quantities. They used citrus seedlings. In their experiments, the amount of the ion absorbed depended on the nature of the other ion of the salt.

It has been noted by some workers, that some chemicals, like calcium, act as antagonistic agents against the entry of some mineral salts into the plant cells. For instance, Hoagland and Davis (1925) found that one ion may retard or accelerate the absorption of another one. Osterhout (1919) and (1922), Raber (1920), Loeb (1912), McCool (1913), Hansteen (1910), Brenner (1925) and Hawkins (1913), mention many experimental evidences for the presence of antagonistic effects possessed by some ions. Scrath and Lloyd (1930) in reviewing the mechanism of the mutual antagonism of mono- and polyvalent cations (like sodium and calcium) regard as due to their opposite effects on permeability. They remark that the antagonism of calcium or other non-toxic cations to the action of low concentrations of more toxic ions of whatever valency, may be the result of the former being supplied in sufficient concentration to lower permeability and keep the others out.

Jacobs (1925) says that ethyl alcohol enters cells with great ease. As to acids and bases, he mentions, that since they are very toxic due to very marked degree of physiological activity of H^+ and OH^- respectively, they are injurious to cells. Similar views have been held by Bayliss (1927).

Recently Cooper, Dorcas and Osterhout (1929) have studied the penetration of strong electrolytes in valonia. They found that the electrolytes employed by them are absorbed very slowly unless the cells are injured.

Orton (1927) measured the permeability of corn seed coats to mercury compounds. He used 4 varieties of sweet and 3 of dent corn. He employed mercuric chloride, chlorophenol mercury sulphate and cyan-cresol mercury. He concludes that the portion of the seed covering the embryo side of the seed is more slowly permeable than covering the endosperm. The rate, according to him, varies with the variety.

From the review of literature it seems safe to infer that in spite of the vast amount of work done along this line, there does not seem to be any great uniformity in the conclusions held at present.

III. Materials and Methods.

A few healthy, large and uniform, ears of Shawnee white variety were selected from the Farmers' Union Cooperative Business Association, Saint Marys, Kansas. Grains from the tip and the butt ends were discarded, because they may be unevenly developed or the endosperm may vary as to the hardness. Since it has been shown by Alberts (1927) that there is a relation of corn endosperm character to absorption of hygroscopic moisture, such a procedure in selecting uniform seeds, from the middle portion of the ear only, seems most desirable. The fuzz of the seeds was removed and they were weighed up to the fourth decimal place in groups of 10 each.

Pyrex test tubes 8 inches in length and one inch in diameter, were cleaned. Their necks were tied with wire. The seeds and the respective solutions were poured in the test tubes, which were labelled and suspended in a water bath, whose temperature was maintained at 25°C. DeKhotinsky electrically heated and regulated theremostat bath was used. It was provided with turbine stirrer for keeping uniform temperature at all levels of the bath. At no time a difference of more than 0.2° centigrade was noted.

The seeds were immersed in various solutions for one hour, at the end of which the liquids were poured in graduated cylinders. The superficial moisture of the seeds was removed by filter paper, without pressing them. They were immediately transferred to weighing bottles, weighed, net increase recorded, the seeds as well as the solution put back in the respective test tubes and suspended in the bath as before. After recording each weight, the weighing bottle was air dried and weighed.

The procedure similar to the one described above was repeated at the end of 2, 4, 8, 16, 32 and 64 hours. There was no particular significanee in selecting these hours, except perhaps, that they formed feasible units of time in geometrical proportion. They were short in the early stages, yet they were long enough in the later limits. Since it was proposed to follow permeability for somewhat longer time, the choice of such intervals was assumed to be satisfactory for this work.

From the increase obtained, percentages were calculated on the basis of original seed weights.

IV. Discussion of Data

The table shows the permeability of water or water and solutes (the latter undetermined) from various solutions in corn seeds held at 25°C. The solutions and their concentrations have been recorded in column II. By referring to the figures recorded in column V, it will be seen that the percentage increase, based on the air dry seed weights, was almost double in B than in A. All samples of corn A and B, expressed similar differential tendency of increase with almost all the solutions (a few exceptions may be seen from the data). It seems that the endosperm character, individual membrane or some other unknown factor, may be responsible for more increase in favor of B at least during the earlier stages of permeability. Later such a variation comes to a minimum and may at times fall within the range of an experimental error. Figures from A and B represent averages of 6 individual readings. Their averages have also been shown in the table.

The average figures shown in column V, indieate that ethyl alcohol penetrated the least (1.87%), M/10 and M/20 sodium hydroxide the most 28.54 and 24.20% respectively). When a small amount of calcium oxide (one-twentieth of the weight of sodium hydroxide) was added to M/10 sodium hydroxide solution, the permeability decreased to less than half. In other words, the presence of calcium oxide inhabited the penetration of M/10 sodium hydroxide at the end of one hour. Similar phenomenon was observed even at the eud of 64 hours. Since several authors have mentioned that calcium acts as antagonistic agent, this study seems to suggest that the antagonism may be due to (1) slowing the rate of absorption, (2) neturalizing the effect of sodium by replacing Na by Ca or (3) by both the factors.

The percentage increase of absorption, at the end of one hour as shown by other solutions (column V) varied but little. Their percentages increase ranged from 8.0 to about 10%. Individual intake may be referred to from the figures presented in column V.

The per cent absorption increase after 1 hour increased very little as shown by other solutions (col. V). This percentage increase ranged from 8.0 to about 10.0. Individual intake may be seen from the figures in col. V.

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Net and percentage absorptions, after 2 hours, have been noted in columns VI and VII. The data seems to show that the percentages are minimum for the alcohol, maximum for M/10 and M/20 NaOH, that is, their tendency is similar to that which was observed at the end of an hour. The effect of CaO is pretty much the same as described before. Of the other solutions, percentage increase for M/10 Nacl was slightly more than for M/20 $NaHSo_4$, caffeine and CH_3 CooH were absorbed about the same within themselves, although different solutions showed variable percentage of permeability. For instance, where M/10 and M/20 CH_3 CooH showed 10.61 to 11.75%, M/10 and M/20 Caffeine 11.76 to 11.96 and M/10 and M/20 Na HSo_4 made 13.24 to 13.26 percent increase. The former two solutions seem to show a closer increase of permeability at the end of two hours. The percentage figures for redistilled water were 12.71.

On comparing the percentage increase between 1 and 2 hours (columns V and VII), in general, a difference to 35 to 50 per cent was noted. However, there was a decided variability between various solutions. The individual increase and their specific differentia may be ascertained from the table (columns V and VII).

Absorption at the end of four hours can be seen in columns VIII and IX. On the average, the trend is practically the same as has been noted already for various solutions. There seems to be an increase in the rate of permeability. The difference in the percentage increase of weight between the end of the first and the fourth hour, on the whole, is about 100 per cent for the individual concentration of the solutions used. Ethyl alcohol seems to be an exception in this respect, since it shows but a very slight decrease instead of any increase.

Table—Permeability of Water and Solutions

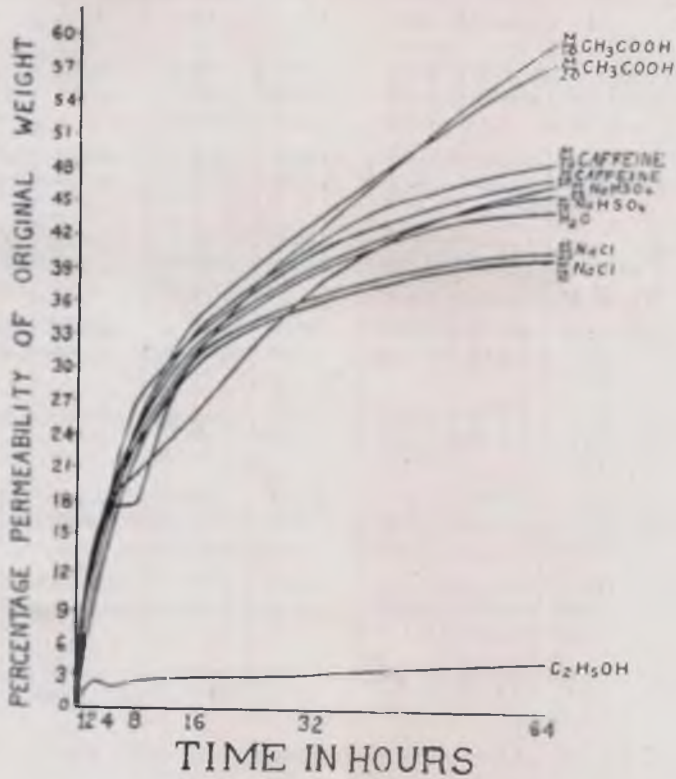
Serial No.	Kind of solution and concentration	Weight of dry seeds	Net weight increase in grams as					
			1 Hour.		2 Hours.		4 Hours.	
			Net.	Per cent.	Net.	Per cent	Net.	Per cent.
1	2	3	4	5	6	7	8	9
1.	Redistilled water.	a 5.2721 b 0.518 Aver	0.3566 0 5188	6.75 12 23	0 5007 0.6758	9.49 15 94	0.7728 0.8843	14.65 20.86
2.	100 % Ethyl Alcohol.	a 5.0751 b 4.2686 Aver	0 0652 0.1057	1.28 2.47 1.87	0.0755 0.1255	1.48 2.93 2.20	0.0604 0.1213	1.10 2.84 2,01
3.	M/10 Sodium Chloride.	a 5.1294 b 4.1108 Aver	0.3384 0.5373	6.59 13,07 9 83	0.4709 0.7057	9.19 17.21 13.20	0.7099 0.9313	13 83 22 65 18.24
4.	M/20 Sodium Chloride.	a 5.1505 b 4.3233 Aver	0.3314 0.4776	6 43 11.05 8.74	0·4921 0.6510	9.55 15.05 12.30	0.7291 0.8316	14.01 19 23 16.62
5.	M/10 Sodium Bisulphate.	a 5.0998 b 4.0481 Aver	0.3575 0.5269	7,01 13.01 10.01	0 4990 0 6762	9.78 16 70 13.24	0.7350 0.8940	14.43 22.08 18.25
6.	M/20 Sodium Bisulphate.	a 5.1847 b 4 1498 Aver	0.3442 0 5943	6 63 14.32 10.47	0.4691 0.7256	9.05 17.48 13.26	0.7295 0 9266	14.07 22.32 18 19
7.	M/10 Sodium Hydroxide.	a 5.3048 b 5 1863 A 5.1888	1.4588 1.5351	27.49 29 60 28.54	2.0788 2.0838	39.18 40.18 39.68	3.1525 3.2207	59.42 62.10 60.76
8.	M/20 Sodium Hydroxide.	a 5.1868 b 5.1837 Aver	1.1219 1.3882	.62 26.78 24.20	1.5725 2.0434	30.31 39.42 34.86	2.3213 2.9397	44.75 56.71 50.73
9.	M/10 Caffeine.	a 5.0829 b 3.2498 Aver	0.3664 0.2871	7.20 8.83 8.01	0.5477 0.4144	10.77 12 75 11.76	0.8091 0 6864	15.92 21.12 18.52
10.	M/20 Sodium Caffeine.	a 5.4208 b 3.2091 Aver	0 3693 0.3102	6.81 9.65 8.23	0.5849 0.4220	10.78 13 15 11.96	0.9138 0.6046	16.85 18.84 17.89
11.	M/10 Sod. Hydroxide and Calcium Oxide.	a 5.1330 b 2.0849 Aver	0.5551 0 4390	10.81 14.23 12.52	0.8497 0.6278	16.55 20.35 18.45	1.2149 0.8693	23 66 28.18 25 92
12.	M/10 Acetic Acid.	a 5.2671 b 3.3518 Aver	0.3191 0 3023	6.05 9.01 7.53	0.4620 0.4175	8.77 12.45 10.61	0.6798 0 5726	12.91 17.08 14.99
13.	M/20 Acetic Acid.	a 4.9136 b 3.3759 Aver	0.3460 0.3185	7.04 9.14 8.09	0.4944 0.4543	10 06 13.45 11.75	0 7893 0.6473	16.06 19.17 17 61

in Zea Mays Seeds at 25° Centigrade

WELL AS IN PERCENTAGES AFTER.

8 HOURS		16 HOURS		32 HOURS		64 HOURS	
NET	PER CENT	NET	PER CENT	NET	PER CENT	NET	PER CENT
10	11	12	13	14	15	16	17
1.1342	21.51	1.6117	30.57	2.0184	38.47	2.4137	45.78
1.1483	27.09	1.4736	34.76	1.6969	40.03	1.9289	45.50
	24.30		32.66		39.25		45.64
0.0730	1.43	0.1008	1.98	0.1245	2.45	0.1515	2.98
0.1578	3.69	0.1678	3.93	0.1860	4.35	0.2712	6.35
	2.56		2.95		3.40		4.66
0.4592	8.95	1 4972	29.13	1.8312	35.70	2.0986	40.91
1.1491	27.95	1.4336	34.87	1.5892	38.66	1.7337	42.17
	18.45		32.00		37.18		41.54
1.0784	20.93	1.5179	29.46	1.8760	36.42	2.1559	41.85
1.1088	25 64	1.4372	33.24	1.6363	37.84	1.7884	41.36
	23.28		31.35		37.13		41.60
1.0867	21.32	1.5126	29.66	1.8468	36.21	2.2034	43.20
1.1126	27.48	1.4539	35.91	1.7317	42.77	2.0573	50.81
	24.40		32.78		39.49		47.00
1.1048	21.30	1.5623	30.13	1.9377	37.37	2.3925	46.23
1.1113	26.77	1 4261	24 36	1.6830	40.55	1.9970	48.12
	24.03		27.24		38 96		47.17
6.3763	120.19	11.3017	211.06	...			...
7.7161	148.80	12.3228	237.60	...	SEEDS DECOMPOSED		...
	134.50		224.33				
3.1254	60.26	8.3861	151.16	...			...
4.0485	78.10	8.8076	169.91	...	SEEDS DECOMPOSED		...
	69.18		160.53				
1.1704	23.02	1.5947	31.37	1.9508	38.37	2.4473	48 16
0.9352	28.77	1.2541	38.59	1.4987	46.11	1.6823	51.76
	25.89		34.98		42.24		49.96
1.3606	25.09	1.8079	31.50	2.1860	40.30	2.5918	47.81
0.8834	27.52	1.1988	37.35	1.4062	43.81	1.5965	49.74
	26.30		34.42		42.05		48.82
1.7173	33.45	2.3964	46.68	2.9709	57.87	3.4181	66.59
1.2596	40.83	1.7840	57.83	2.2407	72.63	2.5757	83.49
	37.14		52.25		65.25		75.04
0.9947	18.88	1.4987	28.45	2.0725	39.35	2.7723	52.63
0.8810	26.28	1.2225	36.47	1.5886	47.32	2.2976	68.55
	22.58		32.46		43.33		60.59
1.1574	23.55	1.6253	33.07	2.1206	43.15	2.9096	59.21
0.9435	27.95	1.2764	37.84	1 5807	46.76	1.9852	58.80
	25.75		35.45		44 95		59.00

At the fourth hour, seeds immersed in M 10 and M 20 NaOH solutions seemed to have been injured because some exmosis took place. The external media changed to yellow color. This was assumed to be due to the corn pigments, which were excreted due to the injurious influence exerted by these solutions. In other solutions (including M 10 NaOH plus CaO) such a change of color was not evident.

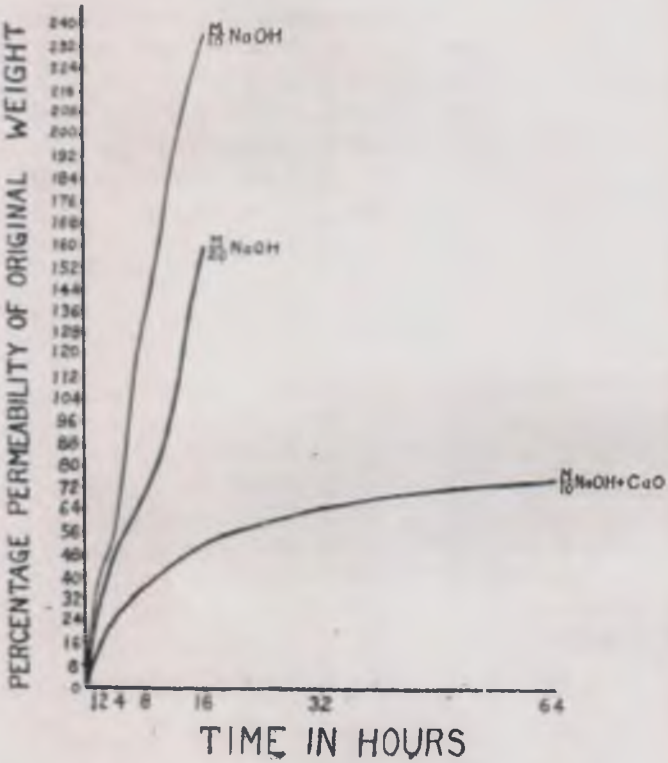


GRAPH I.

Permeability by Zea Mays seeds immersed in solutions of various concentrations

Specific average figures for the absorption of all the solutions at the end of four hours, may be seen in the table. Graph Nos. I and II illustrate the rate of increase at this hour as well as during all the later periods employed in this experiment.

Data presented in columns X and XI show permeability at the close of the eighth hour. The intake has increased for all solutions. With the exception of ethyl alcohol, NaOH. NaOH + Cao and M/10 Nacl the percentage increase on an average, ranged from 22.58 to 26.30%, although the majority of the solutions fell within 24 to 25%. The individual absorption may be verified by referring to the figures. There is a great increase in NaOH. The inhibitory influence of Cao is also distinctly apparent (refer to Graph II).



GRAPH II.

Permeability by Zea Mays seeds immersed in M/10, M/20 Na OH solutions and M/10 plus Cao.

On the whole, solutions with lower concentrations are absorbed more than those with high concentrations. This does not hold true in all cases, because some solutions show tendency just the opposite. This can be well substantiated from the graphs. It seems to the writer, that the relative rate of absorption from lower or higher concentrated solutions, depends in a large measure, on the type of the solute used. No generalization can be made in this respect.

Seeds immersed in NaOH swelled enormously. They were decidedly injured. This may account for their unusual percentage of increase. Calcium oxide showed protective effect even at this period (after 8 hours).

Permeability at the close of 16 hours has been presented in columns XII and XIII. The intake was constantly increasing at this hour. The percentage increment for H_2O, Nacl, $NaHSO_4$, caffeine, NaOH + Cao and CH_3 CooH, was somewhat less than double over the gain made at the end of 4 hours. Ethyl alcohol and NaOH showed exception, because the former increased but very little and the latter increased without any relationship. At this hour there seems to be a distinct difference in the total intake of the solutions, as can be seen from the graphs. For the individual total increase, figures can be referred to.

NaOH solutions decidedly injured the seeds which at this time were entirely decomposed. Their further employment was impossible and unprofitable. Hence they were discarded. On the other hand, similar solution with Cao did not show any adverse effect.

The data obtained for permeability after 32 hours has been shown in columns XIV and XV. The essential points brought out by the figures are practically the same as have been discussed previously, except that there was more total and percentage increase than at any previous hour. It must be noted that up to this period, the percentage increase has been more in favor of M 10 CH_3 CooH than M 20 CH_3 CooH, while after 32 hours this relationship seems to have been reversed (refer to Graph I).

Figures for 64 hours have been recorded in columns XVI and XVII. They show, in the main, about the same tendency as has been indicated by the data for permeability at the smaller periods. Total increase is maximum at this hour. The concentrations of various solutions used in this study, do not seem to have shown any tendency for decrease in the rate of permeability. May be that the seeds were not injured. If there was any injury, certainly it could not be detected by this method. At least there was no visible sign of injury to the seeds, except for those immersed in NaOH and M 10 CH_3 CooH.

At the close of 64 hours, the percentage increase of permeability (based on the original weights of the seeds) seems to run in the following order from maximum to minimum (refer to graph I). NaOH and NaOH plus Cao have not been included, since they have shown somewhat different tendency. (Refer to Graph II):—

M/10 CH_3 CooH, M/20 CH_3 CooH, M/10 Caffeine, M/20 Caffeine, M/20 Na HSO_4, M/10 $NaHSO_4$, H_2O, M/20 Nacl and M/10 Nacl.

V. Summary.

1. Permeability of water, 100 per cent C_2H_5OH, M/10 and M/20, Nacl, $NaHSO_4$, Caffeine, NaOH, NaOH + Cao, CH_3 CooH in Zea Mays seeds at 25°C. has been determined by weight increase method.

2. The data seem to show that after 64 hours these liquids are absorbed from maximum to minimum in the following order: M/10 NaOH, M/20 NaOH, M/10 NaOH + Cao, M/10 CH_3 CooH, M/20 CH_3 CooH, M/10 Caffeine, M/20 Caffeine, M/20 $NaHSO_4$, M/10 $NaHSO_4$, H_2O, M/20 Nacl, M/10 Nacl, and C_2H_5OH.

3. The rate and the increase of periodic permeability have been shown by the data. It seems that various solutions are absorbed at different rates.

4. On the whole, less concentrated are more permeable than more concentrated ones. Exceptions to this phenomenon have been discussed.

5. Calcium has distinctly shown its ability as an antagonistic agent against NaOH injury. It seems from the data that such an influence as has been exerted by calcium, may be due to its inhibition of the rapid entrance of sodium ion.

6. The data bring forth some other points, which have been discussed in the text.

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