PHYSIOLOGICAL STUDIES ON THE GERMINATION OF YEZO SPRUCE SEED

By

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(With 26 Text-figures)

Contents

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I Introduction

All problems of the seed, may ultimately come to its morphological, anatomical, componental, and physiological characters. Numerous studies on the storage and germination of seeds have been made in the field of agricultural science with the purpose of promoting the production and the utilisation. Concerning the forest seeds, however, in recent years no definite progress has been noticed in the problem of relations between the germination and outer factors and of metabolic phenomenon during the germination of the seeds. Under these circumstances physiological studies on the germination of seeds of *Picea jezoensis* and *Picea Glehnii*, which are important forest resources abundantly distributed in all provinces in Hokkaido and Saghalien, have not been reported up to the present time. The writer believes in the urgent necessity of adequate and fundamental on the important forest seeds, and he has attempted to attack the problem from the physiological point of view in the present paper.

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II Materials and Method

As experimental materials the seeds of Yezo spruces, *Picea Glehnii* (Akayezo) and *Picea jezoensis* (Kuroyezo) were used. These seeds were collected at the end of September 1938 in the Teshio and Uriu Experimental Forests of the Faculty of Agriculture, in Hokkaido. The collected seeds were preserved at $-15°C$, in the constantly low temperature laboratory and they were brought out for experiments as necessary. The low temperature laboratory belonging to the Faculty of Science, Hokkaido Imperial University, contains two cold chambers of $-15°C$ and $-35°C$. The experimental procedures of the germination test, the methods of determining the water absorption by seeds, the degrees of their swelling, the permeability of the seed coat, and the osmotic value of seeds and seedlings, or the methods of quantitative micro-analysis of electrolytes and non-electrolytes are described in detail concerning the respective experiments in the following chapters.

III Factors influencing water absorption by seed during germination

It is a matter of common opinion that ordinary mature seeds absorb water in the first stage of their germination and then germinate on a moist substratum, that is to say, the supply of water is an important factor in germination. In general, it is considered that the seeds absorb water, become swollen and then various material changes and other consequent physiological processes occur in the seeds. In other words, the first phases of germination of seed can not occur without water. The water necessary for germination enters into the inner part of a seed through the seed coat and then is absorbed by endosperm and embryo. The processes of absorption of water by living seeds differ of course from those by non-living materials.

The absorption of water by various seeds has been reported by many authors[1] using such agricultural seeds as corn, soy bean, barley, wheat, rye, rape, pea, watermelon etc. These authors are of the opinion that the absorption of water by seeds is markedly influenced by the surrounding

1) ATKINS (1909), BRENCHLEY and WORLEY (1912), BROWN and WORLEY (1912), SHULL (1913), EILTER (1914), RIPPEL (1918), HARRINGTON and CROCKER (1923), SHULL and SHULL, S. P. (1924), DUNGEN (1924), RUDOLFS (1925), OHGA (1926), WOLFE (1926), JONES (1928), BROWN (1931), KISSER and SCHMID (1934).

factors such as temperature, by time, and by structure of the seeds, both morphological and anatomical. For instance, BRENCHLEY and WORLEY (1912), BROWN and WORLEY (1912), and SHULL, S. P. (1924) ascertained that temperature influences the absorption of water by seed and the temperature coefficient *varies with the character of seed, especially the semi-permeability of seed coat.* BROWN (1909) and SHULL (1939) confirmed that the absorption of water by seed is greater than absorption of other substances and WOLFE (1926) proved that the absorption of water by barley grains is influenced by the species and concentration of salts in solutions in which seeds have been soaked. BROWN (1631) recognized that the pale and the cuticular membranes of the seed have a retarding effect of water absorption by seed.

From these investigations it may be seen that surrounding factors and the character of seeds both have a close relation with water absorption by seed, so far as seeds are living or germinating. In the following experiments the writer attempted to investigate the relation between the water absorption by seed of Yezo spruce and the salts, non-electrolytes, hydrogen-ion concentration and temperature, in order to discover the significance of surrounding factors in respect to the water absorption by seeds and the significance of the water absorption in the seed germination.

The amount of absorbed water by seed was determind by measuring with torsion balance the increased weight of seed after they had been placed in the PETRI-dishes, which were kept in the thermostat for a definite number of days. But it is obvious that such a method cannot afford strictly, absolute values of the amount of water absorbed by seed, because the seeds are able to absorb both water and salts dissolved in it. However the increased weight of seed determined by the above stated method may be regarded as practically the value of the weight of water absorbed by seed, for the reason that water is more absorbed by seed from pure water than from salt solutions [BROWN (1909), STILES (1924), and WOLFE (1926)], and from the writer's below described experimental results (Table 1 and Table 2).

The absorption of ammonia and chlorine by the seeds of Yezo spruce

Experiment 1

This preliminary experiment was conducted to prove whether the increased weight of seed in salt solution during germination was directly due to the absorption of water or due to the absorption of salts by seeds. It is reported by MEVIUS (1928), MEVIUS and ENGEL (1929), PIRSCHL (1929), YAMAGUCHI (1929), and LOO (1931), that the absorption of ammonia by the root system of higher plants in a solution containing single ammonium salts as the source of nitrogen or in a mixed solution containing various salts is predominant over that of other cations such as potassium, sodium, magnesium, etc. On the other hand, in this paper various chloride-salts were used as electrolytes. So that in the case of comparison of the amount of absorbed water and the absorbed salts by seeds of Yezo spruce, ammonium chloride was selected in this experiment as the most suitable salt for the above mentioned reason. Experiment was carried out as follows: 200 seeds were submerged in 20 cc of the salt solution in an Erlenmyer's flask having a capacity of 50 cc: after keeping this vessel for a definite number of hours in the thermostat of 20°C., the absorption of ammonia and chlorine was determined by micro-analysis of the external solution.

Determination of ammonia.—Ammonia was estimated by the micro-kjeldahl method. A suitable amount of the sample was placed in a 100 cc long-necked, round-bottomed micro-kjeldahl flask and 2–4 cc of standard hydrochloric acid (0.01 N) was placed in a 60 cc receiving flask for the fixation of the ammonia which is evolved, using a 2 cc micro-burette graduated to 0.01 cc and coloured with one drop of 0.05 percent sodium alizarin sulphonate as indicator which is introduced by means of a glass thread. The point of condenser tube should be slightly below the surface of the acid. Five cc of saturated caustic potash were added to the sample through a separate funnel. If the sample exceeds 20 cc, it is desired to heat the micro-kjeldahl flask over a burner to boiling, in order to hasten the distillation of ammonia. But when the heating was strong and steam powerful, the distillation usually lasted eight minutes. At the end of distillation the receiving flask must be lowered a little, allowing the outlet tube of the cooler to extend above the surface of the acid, so that the inner side of the outlet tube may be rinsed by the water stream. The last operation is of great importance. If the apparatus is disconnected before the lowering of the receiving flask, part of the liquid in the receiving flask would remain in the tube. Moreover, it is advisable to lower the receiving flask and let the water stream pass through the tube usually for two or three minutes, then wash the outside of the tube with redistilled water. When distillation was complete, the excess of HCl was titrated with 0.01 N NaOH, using a 2 cc micro-burette graduated to 0.01 cc. The standard NaOH was carefully prepared according to the direction of CLARK (1928,

p. 195), and stored in a non-alkaline glass bottle, to which a micro-burette, made of glass of the same quality, and soda-lime guard tubes were attached. The standard HCl was prepared using the standard alkali solution.

Determination of chlorine.—Chlorine was estimated using silver nitrate, and ammonium thiocyanate by the method of RUSZNYAK (1921). A suitable amount of the sample was transferred into 100 cc conical flask by means of a micro-pipette and a little excess amount of silver was titrated with ammonium thyocyanate using a 2 cc micro-burette graduate to 0.01 cc using two drops of saturated iron and ammonium sulphate as indicator which is introduced by means of a glass tube.

The analytical results of this Experiment I are shown in Tables I and 2.

Table I

Absorption of NH_3 and Cl by *Picea Glehnii* seeds during 2 days.
Duration of experiment; Apr. 10–20. 1940.

Concentration of NH_4Cl (mol)	No.	Weight of 200 seeds (gr.)	Per 200 seeds		Per 1 seed		Average weight of 1 seed (gr.)
			NH_3 (mg)	Cl (mg)	NH_3 (mg)	C (mg)	
			±0.86	±1.78	±0.004	±0.008	
0.50	1	0.6080	+2.560	+14.180	+0.012	+0.070	0.0030
	2	0.5880	+5.100	+5.320	+0.026	+0.026	0.0029
	3	0.6340	−5.100	+5.320	−0.026	+0.026	0.0032
	Ave.	0.6100	+2.560	+8.280	+0.012	+0.040	0.0031
			±0.68	±1.42	±0.004	±0.006	
0.20	1	0.6340	−8.160	−2.840	−0.040	−0.014	0.0032
	2	0.6480	+6.120	−2.840	+0.030	−0.014	0.0032
	3	0.6070	+8.160	−1.420	+0.040	−0.006	0.0030
	Ave.	0.6297	+6.120	−2.360	+0.030	−0.012	0.0032
			±0.34	+0.70	±0.002	±0.002	
0.10	1	0.5900	± 0	± 0	± 0	± 0	0.0030
	2	0.6620	+2.040	+2.840	+0.010	+0.014	0.0031
	3	0.5320	+2.380	± 0	+0.012	± 0	0.0027
	Ave.	0.5947	+2.220	+2.840	+0.012	+0.014	0.0030
			±0.172	±0.356	±0.0008	±0.0016	
0.05	1	0.6170	+0.172	+0.356	+0.0008	+0.002	0.0031
	2	0.5840	+2.380	+0.356	+0.012	+0.002	0.0029
	3	0.6240	+2.112	± 0	+0.011	± 0	0.0031
	Ave.	0.6083	+1.588	+0.356	+0.008	+0.002	0.0030
			±0.680	±0.14	±0.0004	±0.0008	
0.01	1	0.5870	+0.400	± 0	+0.002	± 0	0.0029
	2	0.5940	+0.540	± 0	+0.003	± 0	0.0030
	3	0.6980	+1.360	−0.140	+0.007	−0.0008	0.0035
	Ave.	0.6263	+0.78	−0.140	+0.004	−0.0008	0.0031

Table 2

Absorption of NH_3 and Cl by *Picea Glehnii* seeds during 4 days.
Duration of experiment; Apr. 10–20. 1940.

Concentration of NH_4Cl (mol)	No.	Weight of 200 seeds (gr.)	Per 200 seeds		Per 1 seed		Average weight of 1 seed (gr.)
			NH_3 (mg)	Cl (mg)	NH_3 (mg)	Cl (mg)	
			±0.86	±1.78	±0.004	±0.008	
0.50	1	0.4920	± 0	+19.500	± 0	+0.098	0.0025
	2	0.5840	−4.260	+1.780	−0.022	+0.008	0.0029
	3	0.5780	+2.560	−5.320	+0.012	−0.026	0.0029
	Ave.	0.5513	−0.560	+5.320	−0.004	+0.026	0.0028
			±0.68	±1.42	±0.004	±0.006	
0.20	1	0.6240	+9.520	−7.100	+0.048	−0.036	0.0031
	2	0.5490	+8.160	−14.180	+0.040	−0.070	0.0028
	3	0.5920	+11.840	−18.440	+0.038	−0.092	0.0030
	Ave.	0.5883	+11.766	−13.240	+0.042	−0.066	0.0029
			±0.34	±0.7	±0.002	±0.002	
0.10	1	0.6130	+2.380	+2.140	+0.012	+0.010	0.0031
	2	0.5930	+3.060	± 0	+0.016	± 0	0.0030
	3	0.5980	+3.060	+1.420	+0.016	+0.008	0.0030
	Ave.	0.6013	+2.840	+1.780	+0.014	+0.010	0.0030
			±0.172	±0.356	±0.0008	±0.0016	
0.05	1	0.6330	+2.212	+0.708	+0.012	−0.004	0.0032
	2	0.5800	+3.060	± 0	+0.016	± 0	0.0029
	3	0.5220	+2.892	± 0	+0.016	± 0	0.0026
	Ave.	0.5783	+2.720	−0.708	+0.016	−0.004	0.0029
			±0.68	±0.14	±0.0004	±0.0008	
0.01	1	0.5370	+0.480	+0.178	+0.002	+0.0008	0.0027
	2	0.5620	+1.160	−0.178	+0.006	−0.0008	0.0028
	3	0.5250	+0.960	± 0	+0.004	± 0	0.0026
	Ave.	0.5413	+0.860	± 0	+0.004	± 0	0.0027

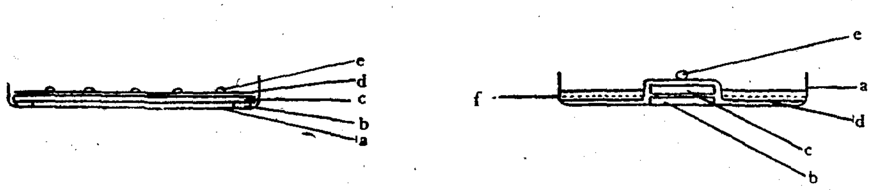
The results shown in Tables 1 and 2 indicate that the absorbed amount of ammonia and chlorine by a seed is very small and its value is, perhaps, within the limit of experimental errors even in the case of ammonia; though the absorbed amount of ammonia is slightly greater than that of chlorine regardless of the concentration of ammonium chloride. At any rate a notable point is that in the results of Experiment 1 the total amount of ammonia and chlorine absorbed by a seed was very small compared to the amount of water absorbed by a seed as described in Experiments 2, 3, …9. Therefore it is natural and right to regard that the increased weight of seed in the salts solution depends upon the absorption of water and does not depend upon the absorption of salts themselves although a small amount of salts, of course, may be absorbed by the seeds in accordance with the concentration of salts. So that from the results of Experiment 1 the writer considered that the absorbed amount of water by seed of Yezo spruce soaked in various solutions may be determinable practically by weighing the increased weight of seeds. From such a point of view the following Experiments 2–12 were carried out.

1 Electrolyte

Experiments 2 and 3 were carried out to investigate the influence of electrolyte on the absorption of water by seed. As electrolytes KCl, NaCl, LiCl, NH_4Cl, $CaCl_2$, $BaCl_2$, and $AlCl_3$ were selected for use. The concentrations of each salt are seen in Table 3 and 4.

The experimental method was as follows: as nearly as possible uniform sized seeds of Yezo spruce were selected as experimental materials. The seeds were placed on a sheet of filter paper being spread out to cover the slide glasses which were laid in PETRI-dishes especially made for germination test (Fig. 1).

Fig. 1. a Fig. 1. b



a ...... PETRI-dish b ...... slide glass c ...... slide glass
d ...... filter paper e ...... seed f ...... water

The diameter of each dish was about 10 cm and its depth was about 1.5 cm. The vessels used in this work were made of non-alkaline glass, and they were washed with chrom-bisulphuric acid and then throughly rinsed with tap and distilled water.

After thus prepared germination sets had been kept in the thermostat of 20°C for a definite number of days, the seeds were rolled on fresh filter paper gently to remove as such of the adherent liquid as possible from the surface of the seeds, and they were then weighed using a torsion balance. By such method the increased weight of seed due to the absorption of water was determined.

Experiment 2

Experiment 2 was performed using seeds of *P. Glehnii* and KCl, NaCl, LiCl, NH_4Cl, $MgCl_2$, $CaCl_2$, $BaCl_2$, $AlCl_3$. The duration of the experiment was 2 days and 4 days. Table 3 and Fig. 2 show the results of this experiment.

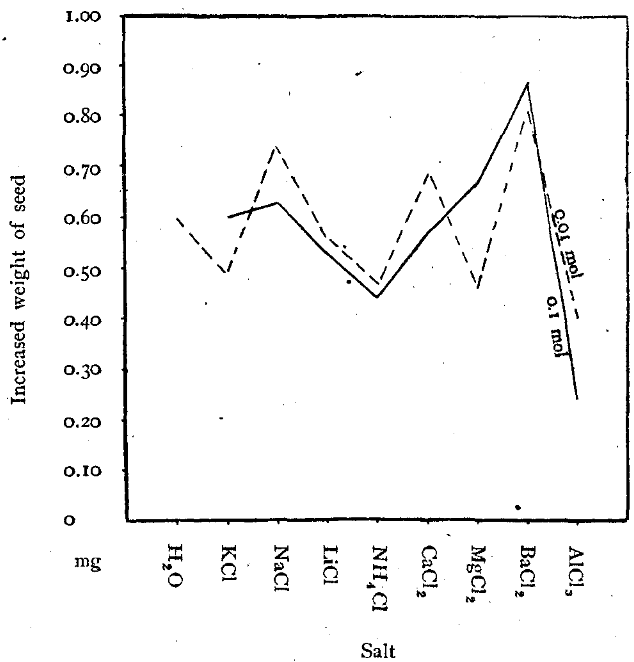
Table 3

Amount of average increased weight of the seeds of *Picea Glehnii* in various salt solutions.
Duration of experiment. Jan. 20–Jan. 31. 1940.

Salt	Concentration (mol)	Average increased weight after (mg)		Average weight of one seed (mg)	Average increased weight per 1 mg of seed (mg)	
		2 days	4 days		2 days	4 days
KCl	0.100 0.010 0.002	0.87 0.74 0.58	1.40 1.17 0.97	3.40 3.96 3.54	0.26 0.19 0.16	0.41 0.29 0.27
NaCl	0.100 0.010 0.002	0.95 0.75 0.63	1.56 1.04 0.98	3.74 3.50 4.00	0.25 0.21 0.16	0.42 0.30 0.25
NH_4Cl	1.00 0.10 0.01	0.43 0.83 1.17	0.76 0.90 1.29	2.89 3.16 3.03	0.15 0.26 0.39	0.26 0.28 0.43
LiCl	0.100 0.010 0.002	1.16 0.99 1.06	1.29 1.13 1.09	3.57 3.10 3.71	0.32 0.32 0.29	0.36 0.36 0.29
$AlCl_3$	0.100 0.010 0.002	0.53 0.50 0.66	0.94 0.98 1.00	3.76 3.34 3.26	0.14 0.15 0.20	0.25 0.29 0.31
$MgCl_2$	0.100 0.010 0.002	0.47 0.74 0.71	0.82 1.00 1.02	3.87 3.90 4.07	0.12 0.19 0.17	0.21 0.26 0.25
$CaCl_2$	0.100 0.010 0.002	0.54 0.50 0.58	1.07 0.75 1.21	3.60 3.41 3.66	0.15 0.15 0.16	0.30 0.22 0.33
$BaCl_2$	0.100 0.010 0.002	0.81 1.11 0.96	1.07 1.26 1.07	3.49 3.16 3.93	0.23 0.35 0.24	0.31 0.40 0.27
H_2O		0.98	1.46	3.80	0.26	0.38

Fig. 2

Comparison of increased weight of the seeds of *Picea Glehnii* after being soaked in various salt solutions for 2 days.



The results of Experiment 2 showed clearly that the absorption of water by the seeds is strongly affected by the kinds and concentration of salts in solution. The absorbed amount of water by seeds of *P. Glehnii* in KCl-, NaCl-, and LiCl-solutions was larger than in the $CaCl_2$-, and $MgCl_2$-solutions. In LiCl-, KCl-, and NaCl-solutions the absorption of water by the seeds was greater in the salts solutions of higher concentrations than in those of lower concentrations. On the contrary the absorption of water in $CaCl_2$-, $MgCl_2$-, and $AlCl_3$-solutions was less in the cases of higher concentrations than of the lower concentrations. For such a manner of absorption of water, the kinds and concentrations of salts may be considered as one of the most responsible determining factors. In order to ascertain the relations between the salts and absorption of water by seed of *P. jezoensis* and to compare them with *P. Glehnii* the following experiment was carried out.

Experiment 3

Experement 3 was performed using *P. jezoensis*; salts and duration of experiment were the same as in Experiment 2. Table 4 and Fig. 3 show the results of Experiment 3.

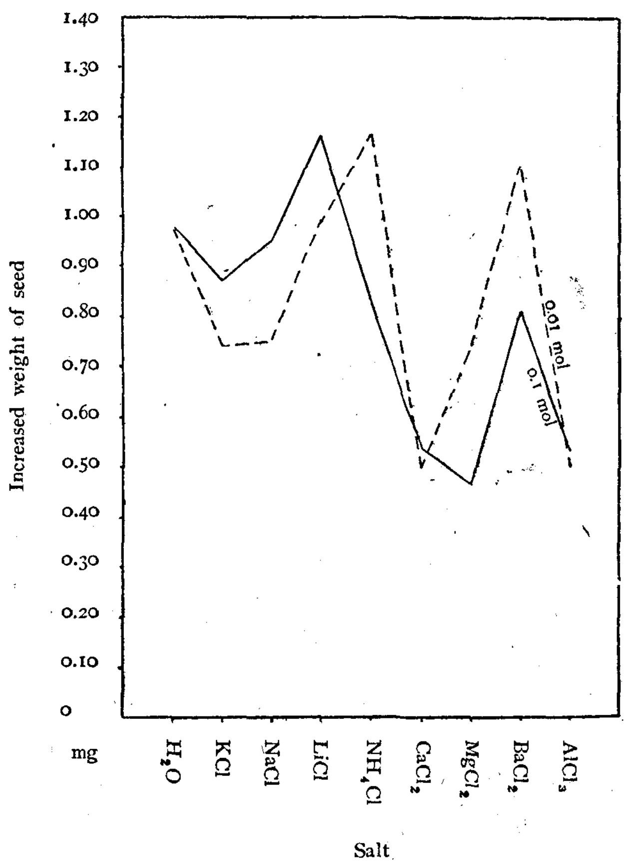
Table 4

Amount of average increased weight of the seeds of *Picea jezoensis* in various salt solutions.
Duration of experiment. Jan. 20–Jan. 31. 1940.

Salt	Concentration (mol)	Average increased weight after (mg) 2 days	4 days	Average weight of one seed (mg)	Average increased weight per 1 mg of seed (mg) 2 days	4 days
KCl	0.100	0.60	0.77	1.49	0.40	0.52
	0.010	0.49	0.84	1.93	0.25	0.44
	0.001	0.63	0.72	2.11	0.30	0.34
NaCl	0.100	0.63	0.82	2.14	0.29	0.38
	0.010	0.74	0.72	1.97	0.38	0.37
	0.002	0.53	0.66	1.90	0.28	0.35
NH_4Cl	1.00	0.27	0.33	1.94	0.14	0.17
	0.10	0.44	0.60	2.11	0.21	0.30
	0.01	0.47	0.60	2.01	0.23	0.30
LiCl	0.100	0.53	0.58	2.56	0.21	0.23
	0.010	0.56	0.69	2.24	0.25	0.31
	0.002	0.54	0.66	2.24	0.24	0.29
$AlCl_3$	0.100	0.24	0.57	1.99	0.12	0.29
	0.010	0.40	0.64	2.27	0.18	0.28
	0.002	0.36	0.65	2.06	0.17	0.32
$MgCl_2$	0.100	0.67	0.91	1.67	0.40	0.54
	0.010	0.46	0.80	1.97	0.23	0.41
	0.002	0.41	0.61	2.01	0.20	0.30
$CaCl_2$	0.100	0.57	0.71	1.97	0.29	0.36
	0.010	0.69	0.73	2.20	0.31	0.33
	0.002	0.54	0.64	2.11	0.26	0.30
$BaCl_2$	0.100	0.86	0.99	2.03	0.42	0.49
	0.010	0.83	0.98	2.09	0.40	0.47
	0.002	0.86	1.00	2.04	0.42	0.49
H_2O		0.60	0.75	2.08	0.29	0.36

Fig. 3

Comparison of increased weight of the seeds of *Picea jezoensis* after being soaked in various salt solutions for 2 days.



From Table 4 it will be seen that the relations between the absorption of water and kinds and concentrations of salts are not so clear as in the case of *P. Glehnii*. What is the cause of this unclearness? First of all, the difference of size and weight of seeds between *P. Glehnii* and *P. jezoensis*. For exampl, the average weight and size of *P. jezoensis* seed is 1.8–2.2 mg and 1.4×2.5 mm respectively, but that of *P. Glehnii* seed is 3.5–4.0 mg and 1.9–3.6 mm. Therefore it happened that the difference of each average increased weight of seed of *P. jezoensis* by absorption of water in various kinds and concentrations of salts is slight, and in extreme instances, the determined difference appeared within the limit of experimental errors.

The works of RUDOLFS (1925), WOLFE (1926) and PRINGSHEIM (1930), who used pea, maize, wheat, lupine and soy beans as experimental materials, indicated clearly the relations between the differences of absorption of water by seed and kinds and concentrations of salts. Those results may be attributable to the large size and weight of seeds or grains which are very large to compare with those of *P. jezoensis* used in the present work. In general, it may be considered that electrolytes which accelerate the swelling of the protoplasm or softening the plasma membrane have a tendency to increase the permeability of seed coat or outer layer of inner seed to water. That is to say, in the above described Experiments 2 and 3, electrolytes such as KCl, NaCl, LiCl, increased more the weight of seed in salt solutions of the higher concentrations than in the lower concentrations. In this case it may be regarded easily from the results of Experiment 1 that the more increased weight of seed in higher concentrations was not due to the absorption of salts themselves but was due to the absorption of water. On the other hand, the electrolytes such as $MgCl_2$, $CaCl_2$ which check or diminish the swelling of the protoplasm and the plasma membrane may have a tendency to decrease the permeability of seed coat to water or salts. In Experiment 2 such salts have decreased the amount of absorption of water in accordance with increasing the concentration of salts because of the decrease of the permeability of the seed coat to water. From the results of Experiments 1 and 2 it may be considered that the differences in absorption of water by seed occur in accordance with the species of seed, especially influenced by its size and weight, and with the kinds and concentrations of salts, more exactly the ionic effects such as those by K, Na, Li, Ca, or Mg ions.

2 Antagonism

It has long been known that solutions of certain single salts are toxic to living organisms placed therein, but that in a mixed solution of these salts the organisms function normally and are not injured. The works of OSTERHOUT (1907, 1908), HANSTEEN-CRANNER (1910), KAHHO (1921), HOAGLAND (1923. a. b.), LUNDEGÅRDH and MORAVÉK (1924), and ILJIN (1927) showed that the presence of one salts influences the physiological function of another salt. In the present case the following experiment was undertaken in order to see if the antagonism between the salts can exist in the case of absorption of water by seeds of *P. Glehnii* and *P. jezoensis*.

Experiment 4

This experiment was carried out using *P. Glehnii* and *P. jezoensis* seeds and the following salts: KCl, NaCl, $MgCl_2$, and $CaCl_2$. The concentration of salts was 0.1 mol, and two salts were mixed in various ratios as following: details are seen in Table 5. $CaCl_2$: KCl, KCl : NaCl, NaCl : $CaCl_2$, $MgCl_2$: KCl, $MgCl_2$: NaCl, $MgCl_2$: $CaCl_2$. The determination of absorbed water by seed was conducted in the manner described above in Experiment 2. Tables 5 and 6 show the data of this experiment.

Table 5

Amount of average increased weight of the seeds of *Picea Glehnii* in mixed salt solutions.

Salts	Average increased weight after (mg)			Average green weight of seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
H_2O	1.1	1.3	1.7	2.5	0.4	0.5	0.7
$CaCl_2$	0.9	1.1	1.3	2.7	0.3	0.4	0.5
Ca(3) : K(2)	1.2	1.3	1.4	2.9	0.4	0.5	0.5
Ca(1) : K(4)	1.5	1.5	1.6	2.4	0.6	0.6	0.7
Ca(1) : K(9)	1.0	1.2	1.4	1.8	0.6	0.7	0.8
KCl	0.8	0.9	1.1	2.8	0.3	0.3	0.4
K(9) : Na(1)	1.1	1.4	1.6	3.4	0.3	0.4	0.5
K(4) : Na(1)	1.4	1.4	1.5	2.9	0.5	0.5	0.5
K(2) : Na(3)	1.5	1.5	1.8	3.5	0.4	0.4	0.5
NaCl	1,9	1.1	1.2	2.9	0.3	0.4	0.4
Na(9) : Ca(1)	1.2	1.4	1.6	2.3	0.5	0.6	0.7
Na(4) : Ca(1)	1.1	1.2	1.3	2.1	0.5	0.6	0.6
Na(2) : Ca(3)	0.4	0.6	0.9	2.9	0.1	0.2	0.3
$MgCl_2$	1.2	1.5	1.6	3.1	0.4	0.5	0.5
Mg(9) : K(1)	1.0	1.2	1.3	2.2	0.5	0.5	0.6
Mg(4) : K(1)	0.9	1.1	1.2	3.0	0.3	0.4	0.4
Mg(2) : K(3)	1.1	1.4	1.4	2.6	0.4	0.5	0.5
Mg(1) : Na(9)	1.2	1.3	1.4	3.5	0.3	0.4	0.4
Mg(1) : Na(4)	1.3	1.3	1.6	2.8	0.4	0.4	0.6
Mg(3) : Na(2)	1.0	1.2	1.4	2.7	0.4	0.4	0.5
Mg(9) : Ca(1)	0.8	1.0	1.2	2.3	0.3	0.4	0.5
Mg(4) : Ca(1)	1.3	1.5	1.6	2.4	0.5	0.6	0.7
Mg(2) : Ca(3)	1.0	1.1	1.3	2.1	0.5	0.5	0.6

Table 6

Amount of average increased weight of the seeds of *Picea jezoensis* in mixed salt solutions.

Salts	Average increased weight after (mg)			Average green weight of seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
H_2O	0.6	0.9	1.2	2.5	0.2	0.4	0.5
$CaCl_2$	0.6	0.8	1.2	2.5	0.2	0.3	0.5
Ca(3) : K(2)	0.7	0.8	1.1	2.1	0.3	0.4	0.5
Ca(1) : K(4)	0.4	0.9	1.1	2.3	0.2	0.4	0.5
Ca(1) : K(9)	0.6	1.1	1.2	2.2	0.3	0.5	0.5
KCl	1.2	1.2	1.5	2.2	0.5	0.5	0.7
K(9) : Na(1)	0.6	0.8	1.0	2.0	0.3	0.4	0.5
K(4) : Na(1)	0.5	0.5	1.1	1.8	0.3	0.3	0.6
K(2) : Na(3)	0.3	0.5	1.2	2.0	0.2	0.3	0.6
NaCl	0.4	0.7	1.4	2.1	0.2	0.3	0.7
Na(9) : Ca(1)	0.7	1.0	1.1	2.3	0.3	0.4	0.5
Na(4) : Ca(1)	0.5	0.7	1.0	2.4	0.2	0.3	0.4
Na(2) : Ca(3)	0.5	0.9	1.1	2.5	0.2	0.4	0.4
$MgCl_2$	1.1	1.5	1.3	1.8	0.6	0.8	0.7
Mg(9) : K(1)	0.7	1.2	1.4	1.9	0.4	0.6	0.7
Mg(4) : K(1)	0.6	1.1	1.3	1.9	0.3	0.6	0.7
Mg(2) : K(3)	0.6	1.0	1.2	1.7	0.4	0.6	0.7
Mg(9) : Na(1)	0.9	1.0	1.3	2.3	0.4	0.4	0.6
Mg(4) : Na(1)	0.6	0.8	1.2	2.0	0.3	0.4	0.6
Mg(2) : Na(3)	0.8	1.2	1.4	2.2	0.4	0.5	0.6
Mg(9) : Ca(1)	0.7	1.2	1.3	2.3	0.3	0.5	0.6
Mg(4) : Ca(1)	0.7	1.1	1.3	1.3	0.5	0.8	1.0
Mg(2) : Ca(3)	0.6	1.0	1.3	1.9	0.3	0.5	0.9

From Tables 5 and 6 it will be seen that the absorption of water by seeds of *P. Glehnii* and *P. jezoensis* is usually more accelerated in the KCl solution than in another salt. The accelerating effect of KCl upon the absorption of water was observed obviously in both seeds in Exp. 3, and KCl was more fovourable to absorption of water than NaCl. The effect of NaCl and $MgCl_2$ on the absorption of water was nearly the same. $CaCl_2$ was worst of all in respect to the acceleration of water absorption but the retarding effect of $CaCl_2$ upon the absorption of water, as observed in the case of *P. Glehnii*, was not obvious in *P. jezoensis*. The order of salts in regard to accelerating the absorptian of water was found as follows: $KCl > MgCl_2 > NaCl > CaCl_2$. As seen in this series KCl and $MgCl_2$

were found to be favourable to the absorption of water. But whenever the experimental material was changed, the favourable effects were often replaced by unfavourable and instead of acceleration, a retardation of water absorption usually took place. The effects of the mixed salt solutions of KCl and $CaCl_2$ on the absorption of water were less than that of KCl, and the more the amount of KCl was mixed in, the less the effects of $CaCl_2$ became. In general, in each mixed salt solution, in the presence of $CaCl_2$ salt the absorption of water was more or less retarded, while in the presence of KCl or NaCl salts the absorption of water was more or less accelerated. From these results of Experiment 3 it may be seen that between the following cations there exists antagonism on the absorption of water: Ca and K, Ca and Na, K and Mg, and that K and Na are not antagonistic to each other. Such an antagonistic action of K and Ca cations has also been observed by BRENNER (1920) and KAHHO (1924) in their studies on the effect of salts on the resistance to the acid of plant protoplasm.[1] HANSTEEN-CRANNER (1919) observed the different effects of K and Ca on the dissolution of phosphatide out of storage tissue of red beet and root system of seedlings. BENECKE (1907) has studied the retarding effect of calcium ion on the entrance of iron into Spirogyra. Their results showed that K furthers the swelling of protoplasm and Ca retards it. In Experiment 4, also, the effects of K and Ca cations on the increasing of seed weight or the absorption of water may be explained on the ground of the difference of the effects of these ions in changing the properties of the seed coat and the seed itself.

3 Non-electrolyte

It is generally well known that the action of non-electrolytes on living organisms differs from that of the electrolytes in respect of the osmotical phenomenon, action of ions or molecules and their specific action[1], for example, special permeability of urea and glycerine. In this section the following Experiments 5–8 were made to investigate the effect of non-electrolytes on the absorption of water and to compare with that of electrolytes observed in Experiment 3 described previously.

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1) DE VRIES (1889), FITTING (1917), HÖFLER and STIEGLER (1921), HÖFLER and WEBER (1926), YAMAGUCHI (1930), BOGEN (1937, 1938).

Experiment 5

Saccharose and seeds of *P. Glehnii* and *P. jezoensis* were used. The method of determination of increased weight of seed and other experimental methods of Experiments 5–8 were the same as in Experiment 2. The results of the present experiment are shown in Tables 7 and 8 and in Fig 4.

Table 7

Amount of average increased weight of the seeds of *Picea Glehnii* in saccharose solution. Temp. 20°C
Duration of experiment: Jan. 11–Feb. 20. 1940.

1) Sep. 12. 1940–Sep. 20. 1940.

Concentration of saccharose (mol)	Average increased weight after (mg)				Average green weight of one seed (mg)	Average increased weight per 1 mg of seed (mg) after			
	2 days	4 days	6 days	8 days		2 days	4 days	6 days	8 days
1.00	0.38	0.64	—	—	3.53	0.11	0.18	—	—
0.80	0.94	0.98	—	—	3.57	0.26	0.27	—	—
0.60	0.84	0.96	—	—	2.87	0 29	0.33	—	—
0.45[1)]	0.56	0.79	1.03	1.10	2.86	0.20	0.28	0.36	0.38
0.40	0.93	1.20	—	—	3.13	0.31	0.38	—	—
0.40[1)]	0.86	1.04	1.21	1.44	2.93	0.29	0 35	0.41	0.49
0.35[1)]	0.86	1.07	1.15	1.20	2.99	0.29	0.36	0 38	0.40
0.20	1.04	1.17	—	—	3.34	0.31	0.35	—	—
0.10	0 90	1.27	—	—	4.01	0.22	0.32	—	—
0.02	1.05	1 30	—	—	3.89	0.27	0.33	—	—
0.01	1.14	1.36	—	—	4.07	0.28	0.33	—	—

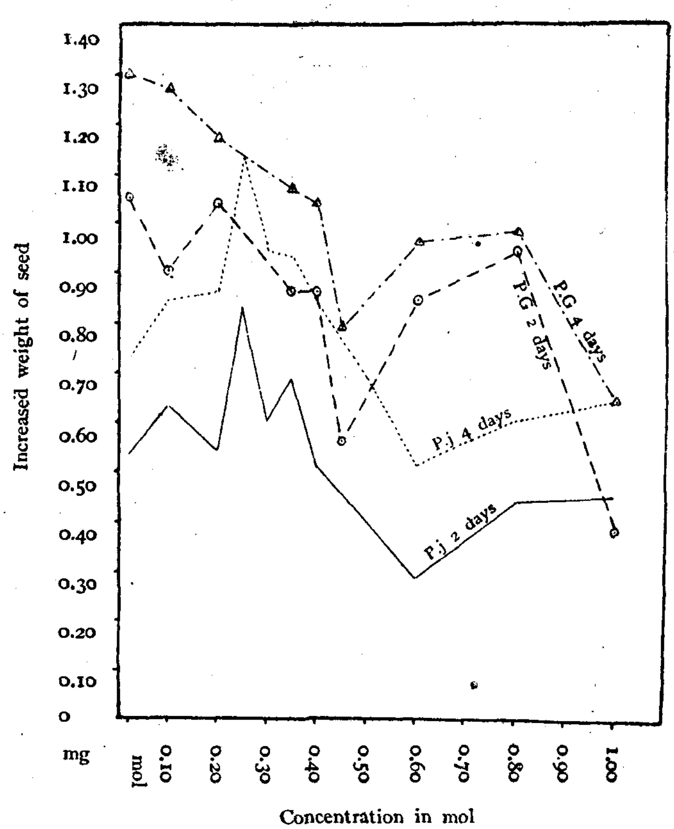
Table 8

Amount of average increased weight of the seeds of *Picea jezoensis* in saccharose solution. Temp. 20°C
Duration of experiment: Jan. 11–Feb. 20. 1940.
1) Sep. 12. 1940–Sep. 20. 1940.

Concentration of saccharose (mol)	Average increased weight after (mg)				Average green weight of one seed(mg)	Average increased weight per 1 mg of seed (mg) after			
	2 days	4 days	6 days	8 days		2 days	4 days	6 days	8 days
1.00	0.45	0.64	—	—	2.00	0.23	0.32	—	—
0.80	0.44	0.60	—	—	2.30	0.19	0.26	—	—
0.60	0.29	0.33	—	—	2.26	0.13	0.15	—	—
0.40	0.51	0.83	—	—	2.07	0.25	0.40	—	—
0.35[1)]	0.69	0.93	1.01	1.06	1.83	0.38	0.51	0.55	0.58
0.30	0.60	0.94	1.06	1.00	1.36	0.44	0.69	0.78	0.74
0.25	0.83	1.13	1.23	1.20	2.36	0.35	0.48	0.52	0.51
0.20	0.54	0.86	—	—	2.01	0.27	0.43	—	—
0.10	0.63	0.84	—	—	2.10	0.30	0.40	—	—
0.02	0.53	0.73	—	—	1.93	0.27	0.38	—	—
0.01	0.85	1.03	—	—	2.03	0.42	0.51	—	—

The results of Experiment 5 (Table 7) show that the increasing of weight of seeds laid on the moistened filter paper with saccharose solution of higher concentrations than 0.4 mol is less than in the solution of lower concentrations. In general, the higher the concentration of saccharose the less the increase of seed weight became. In other words, the absorption of water by the seeds in the saccharose solution of lower concentrations is larger than in the solution of higher concentrations. Such a tendency as stated above is the same in both seeds, *P. Glehnii* and *P. jezoensis*. In these cases, the osmotic phenomenon and permeability of saccharose may be important factors influencing the absorption of water by the seeds in relation to the concentration of saccharose solution. The mechanism of the absorption of water by seed may be related intimately with the suction force of seeds or seedling, and this suction force may govern the germination of seeds. There will be some discussion of these problems in the latter parts of this paper considering the permeability of saccharose and suction force of seedlings.

Fig. 4
Comparison of increased weight of the seeds of *Picea Glehnii* and *Picea jezoensis* after being soaked in saccharose solution of various cancentrations for 2 or 4 days.



Experiment 6

Glycerine and seeds of *P. Glehnii* and *P. jezoensis* were used. Results are shown in Tables 9 and 10 and Fig. 5.

From the tabulated results it may be seen that the increasing of seed weight in the glycerine solution of higher concentrations is less than in the

Table 9

Amount of average increased weight of the seeds of *Picea Glehnii* in glycerine solution. Temp. 20°C. Duration of experiment: Jan. 25–Feb. 2. 1940.

1) Feb. 24–Mar. 3. 1940.

Concentration of glycerine (mol)	Average increased weight after (mg)				Average green weight of one seed(mg)	Average increased weight per 1 mg of seed (mg) after			
	2 days	4 days	6 days	8 days		2 days	4 days	6 days	8 days
1.30	0.98	1.22	1.50	1.70	3.11	0.32	0.39	0.48	0.55
1.20	0.83	0.97	1.13	1.13	2.63	0.32	0.37	0.43	0.43
1.10	0.94	1.16	1.30	1.40	2.83	0.33	0.41	0.46	0.49
1.00	0.91	1.16	1.08	1.16	3.29	0.28	0.35	0.33	0.35
1.00[1)]	1.10	1.30	—	—	3.73	0.29	0.35	—	—
0.80	1.09	1.10	1.22	—	3.47	0.31	0.32	0.35	—
0.60	0.97	1.14	1.40	—	3.89	0.25	0.29	0.36	—
0.40	1.07	1.19	1.40	—	3.64	0.29	0.33	0.38	—
0.20	0.81	1.30	1.35	—	3.89	0.21	0.33	0.35	—
0.10[1)]	1.19	1.39	—	—	3.66	0.32	0.38	—	—
0.01[1)]	1.29	1.43	—	—	3.30	0.39	0.43	—	—
H_2O	1.26	1.43	—	—	3.56	0.35	0.40	—	—

Table 10

Amount of average increased weight of the seeds of *Picea jezoensis* in glycerine solution. Temp. 20°C. Duration of experiment: Jan. 26–Feb. 3. 1940.

1) Feb. 24–Mar. 3. 1940.

Concentration of glycerine (mol)	Average increased weight after (mg)				Average green weight of one seed(mg)	Average increased weight per 1 mg of seed (mg) after			
	2 days	4 days	6 days	8 days		2 days	4 days	6 days	8 days
1.30	0.40	0.70	0.85	1.00	2.09	0.19	0.33	0.41	0.48
1.20	0.54	0.86	0.73	0.80	2.07	0.26	0.42	0.35	0.39

Table 10 (*Continued*)

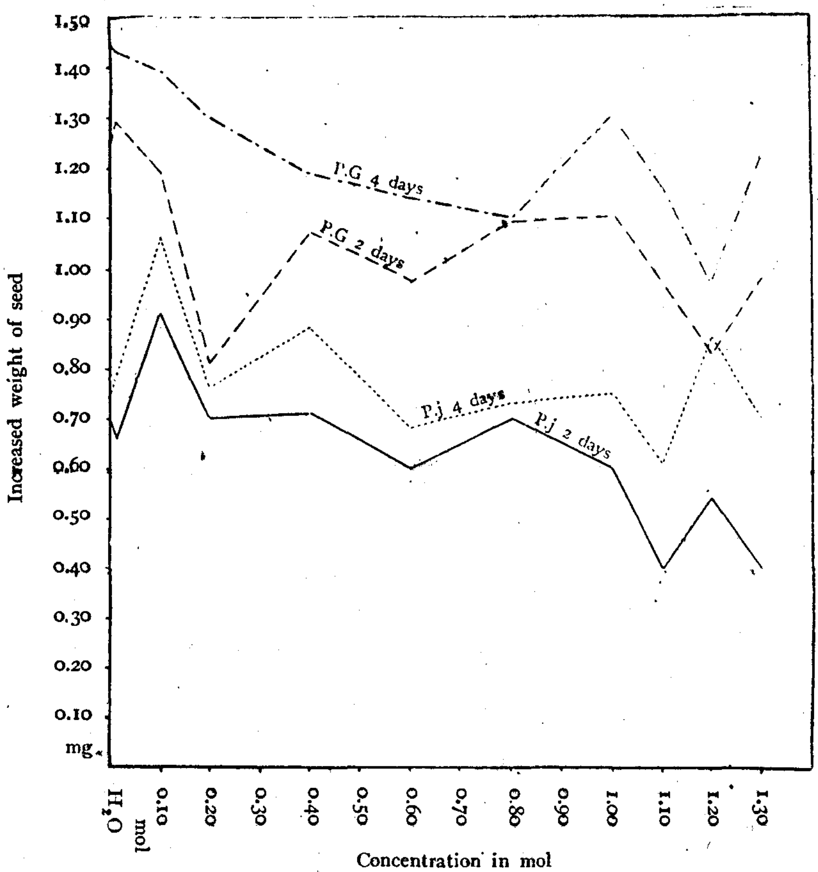
Concentration of glycerine (mol)	Average increased weight after (mg)				Average green weight of one seed (mg)	Average increased weight per 1 mg of seed (mg) after			
	2 days	4 days	6 days	8 days		2 days	4 days	6 days	8 days
1.10	0.40	0.61	0.48	0.60	1.74	0.23	0.35	0.28	0.34
1.00	0.63	1.08	0.93	1.00	1.81	0.35	0.60	0.51	0.55
1.00[1]	0.60	0.75	—	—	1.90	0.31	0.39	—	—
0.80	0.70	0.73	0.88	—	2.21	0.32	0.33	0.40	—
0.60	0.60	0.68	0.78	—	2.46	0.24	0.28	0.32	—
0.40	0.71	0.88	0.98	—	2.17	0.33	0.41	0.45	—
0.20	0.70	0.76	0.88	—	2.19	0.32	0.35	0.40	—
0.10[1]	0.91	1.06	—	—	1.86	0.48	0.57	—	—
0.01[1]	0.66	0.78	—	—	2.26	0.29	0.35	—	—
H_2O	0.70	0.74	—	—	2.19	0.31	0.33	—	—

lower concentrations, but the difference of increasing of seed weight observed in the solutions of higher concentrations and in those of lower ones is less than that in the solutions of saccharose. This difference in weight increase between glycerine and saccharose, perhaps, may be due to the higher permeability to glycerine.[1] According to FITTING (1919) the concentration of glycerine at incipient plasmolysis is larger than that of saccharose, though its values vary with the species of materials and the differences of experimental methods. That is to say, glycerine more easily permeates the plant cells than saccharose does. Therefore, in the present experiment with glycerine, it may be considered that the seeds may absorb both water and glycerine, though the absorption of water is very large in volume in comparison with that of glycerine. Consequently it may happen that the increasing of seed weight in the glycerine solution of higher concentration was not so small in comparison with that of the lower concentrations as was seen in the saccharose solution. Experiments on the high permeability of glycerine and its physiological rôle on the germination of seed will be reported at a later chapter of this paper.

1) KLEBS (1887), DE VRIES (1888), FITTING (1919), BÄRLUND (1929), SCHMIDT (1936), KREUZ (1941).

Fig. 5

Comparison of increased weight of the seeds of *Picea Glehnii* and *Picea jezoensis* after being soaked in glycerine solution of various concentrations for 2 or 4 days.



Experiment 7

Urea and seeds of *P. Glehnii* and *P. jezoensis* were used. The results are shown in Tables 11 and 12 and Fig. 6.

The results shown in Table 11 and Fig. 6 indicate that the increasing of seed weight of *P. Glehnii* in urea solutions of higher concentrations was decidedly superior to that in the solutions of lower concentrations. This case differs greatly from that of saccharose. The divergence between the results of urea experiment and those of saccharose experiment may depend,

Table 11

Amount of average increased weight of the seeds of *Picea Glehnii* in urea solution. Temp. 20°C.
Duration of experiment: Jan. 10–Jan. 16. 1940.

Concentration of urea (mol)	Average increased weight after (mg) ±0.1		Average weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)	
	2 days	4 days		2 days	4 days
1.00	0.88	1.47	3.59	0.25	0.41
0.80	1.06	1.26	3.02	0.35	0.42
0.60	1.20	1.33	3.24	0.37	0.41
0.40	1.00	1.16	3.71	0.27	0.31
0.20	1.34	1.34	3.97	0.34	0.34
0.10	0.87	1.12	4.03	0.22	0.28
0.02	0.55	0.83	3.79	0.15	0.22

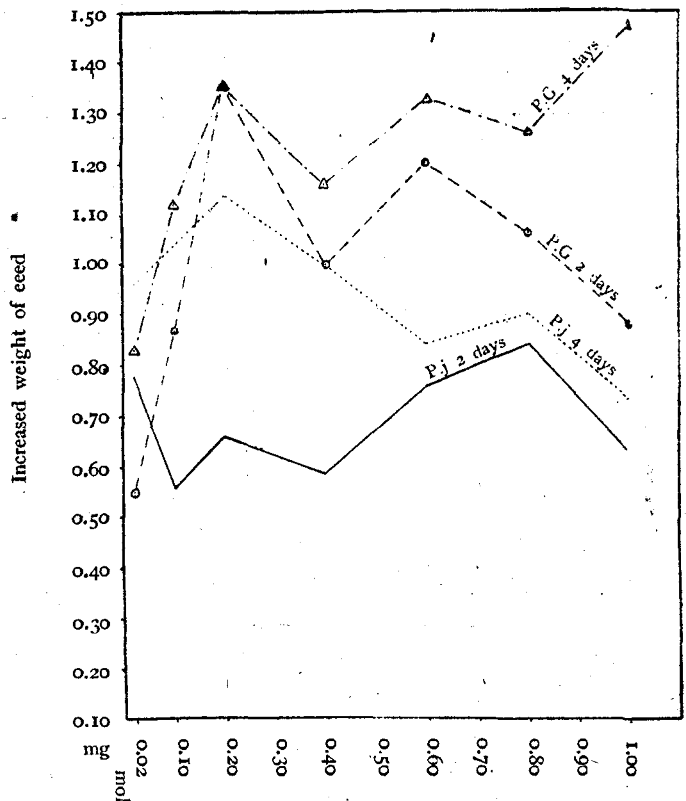
Table 12

Amount of average increased weight of the seeds of *Picea jezoensis* in urea solution. Temp. 20°C.
Duration of experiment: Jan. 10–Jan. 16. 1940.

Concentration of urea (mol)	Average increased weight after (mg) ±0.1		Average weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)	
	2 days	4 days		2 days	4 days
1.00	0.63	0.73	1.89	0.33	0.39
0.80	0.80	0.87	1.81	0.44	0.48
0.60	0.76	0.84	2.20	0.35	0.38
0.40	0.59	1.00	2.14	0.28	0.47
0.20	0.66	1.14	2.07	0.32	0.55
0.10	0.56	0.63	1.87	0.30	0.34
0.02	0.78	0.96	2.00	0.39	0.48

Fig. 6

Comparison of increased weight of the seeds of *Picea Glehnii* and *Picea jezoensis* after being soaked in urea solution of various concentrations for 2 or 4 days.



absolutely, on the physico-chemical difference of urea and saccharose. Of course, urea permeates[1] very easily through the plasma membrane of plant cells. This fact can be demonstrated by the deplasmolytic or plasmolytic method using urea in surrounding solution. Therefore if the seeds are soaked or laid in the urea solutions of various concentrations, it may occur that the seeds absorb both water and urea, and accordingly the increasing

1) DE VRIES (1889), FITTING (1917), HÖFLER and STIEGLER (1921), HÖFLER and WEBER (1926), YAMAGUCHI (1930), HÖFLER (1934), WAHRY (1936), HEIM (1937), BOGEN (1937, 1938), and KREUZ (1941).

of seed weight in urea solutions does not appear in parallel with the increasing or decreasing of the concentration of urea as was seen in the experiment with saccharose. In this case urea may play a rôle in accelerating the water absorption, and the special permeability of urea itself may be regarded as an important factor in the increasing of seed weight in urea solutions of higher concentrations. But, these phenomena were not recognized, so clearly with the seeds of *P. jezoensis* as with those of *P. Glehnii* (Table 12). Possibly it happened because of the small size and weight of the seed of *P. jezoensis* as pointed out already in this paper.

Experiment 8

Alcohol and seeds of *P. Glehnii* and *P. jezoensis* were used. Results are shown in Tables 13 and 14 and Fig. 7.

Table 13

Amount of average increased weight of the seeds of *Picea Glehnii* in alcohol solution. Temp. 20°C. Duration of experiment: Mar. 16–12. 1940.

Concentration of alcohol	Average increased weight after (mg)		Average weight of	Average increased weight per 1 mg of seed after (mg)	
%	2 days	4 days	seed (mg)	2 days	4 days
H_2O	1.00	1.40	3.45	0.29	0.41
1	0.99	1.14	3.29	0.30	0.35
5	0.91	1.06	3.54	0.26	0.30
10	1.01	1.07	3.34	0.30	0.32

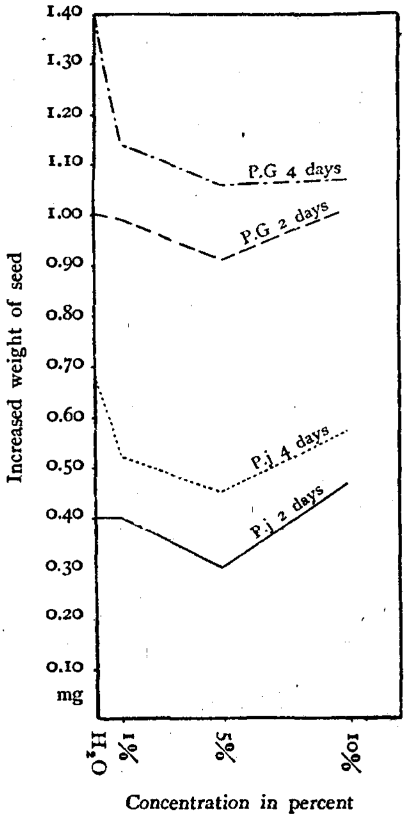
Table 14

Amount of average increased weight of the seeds of *Picea jezoensis* in alcohol solution. Temp. 20°C. Duration of experiment: Mar. 16–22. 1930.

Concentration of alcohol	Average increased weight after (mg)		Average weight of	Average increased weight per 1 mg of seed after (mg)	
%	2 days	4 days	seed (mg)	2 days	4 days
H_2O	0.40	0.56	2.20	0.18	0.25
1	0.40	0.52	2.19	0.18	0.24
5	0.30	0.45	2.46	0.12	0.18
10	0.47	0.57	2.17	0.22	0.26

Fig. 7

Comparison of increased weight of the seeds of *Picea Glehnii* and *Picea jezoensis* after being soaked in alcohol solutions of various concentrations for 2 or 4 days.



The results of Exp. 8 showed no increase of the weight of seeds which were laid on the filter paper moistened with alcohol solution of various concentrations. From these results the question arises whether alcohol is or is not beneficial for the absorption of water complicated by the fact that alcohol itself is very absorbable by seed. In the latter case the specific gravity of alcohol is responsible for the decreasing of seed weight owing to the increasing of alcohol percentage, because the specific gravity of alcohol solution is decreased in accordance with the increasing of the weight of alcohol in alcohol solution. In consequence of this one, or two processes, a decreasing of seed weight may result, because of the special permeability of alcohol for plant cells as reported by TOIVONEN (1934) and RUHLAND, ULRICH and ENDO (1938). Further considerations on these problems will be presented in a later part of the present paper.

4 Hydrogen-ion concentration

Since the fundamental works of SÖRENSEN (1909), MICHAELIS (1909), SMALLS (1929) and others were published it has been ascertained that the concentration of hydrogen-ions is one of the important factors in the regulation of the physiological phenomena of living organisms. Concerning the present problems of the absorption of water by seeds it is highly probable that the hydrogen-ion concentration may play also an important rôle. Experiment 9–14 were intended to see how the hydrogen-ion concentration influences the absorption of water by seed. Solutions of various concentrations of hydrogen-ions were made up using a buffer mixture.

Among several kinds of buffer mixtures, organic and inorganic, sodium phosphate mixture, perhaps, is the most favourable one for biological studies. Loo (1931) made the following statements about it; firstly it is not toxic to living cells as phthalate or acetate are; secondly, both the cation and anion are poorly absorbed by the plant and its effect on the absorption is relatively insignificant as compared to K, Ca and other ions, beside these good points the sodium phosphate mixture possesses tolerable buffer power at comparatively low concentrations and therefore is very suitable for the present purpose. SAKAMURA (1930 a, b) and YAMAGUCHI (1929), also, used this phosphate mixture in their biological studies. For these reasons, a sodium phosphate mixture was employed in the present study. As stock solutions, M/5 H_3PO_4 (MERCK's guaranteed reagent), M/5 NaH_2PO_4 (TAKEDA extra pure) and M/5 Na_2HPO_4 (TAKEDA pro analysis) were prepared; a fixed amount of them was taken with burette and diluted to secure the required pH-value.

Experiment 9

In the present experiment the concentration of phosphate mixture was M/5. The quantity of phosphate in the phosphate mixture of various pH-values was constant, and they were mixed in the following proportions.

pH series	M/5 H_3PO_4 cc.	M/5 NaH_2PO_4 cc.	M/5 Na_2HPO_4 cc.	Obtained pH
3.0	10	90.0	0	3.0
4.0	1.0	99.0	0	4.0
5.0	0	97.0	3.0	5.0
6.0	0	70.0	30.0	6.0
7.0	0	24.0	76.0	7.0
8.0	0	2.0	98.0	8.0
9.0	0	0	100	8.8

Results are shown in Tables 15 and 16.

Table 15

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. PO_4–C,[1] M/5.[2] Duration of experiment: Dec. 1–6. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
H_2O	0.7	0.9	1.4	4.0	0.2	0.2	0.4
3.0	0.5	0.9	1.4	3.3	0.1	0.2	0.4
4.0	0.6	1.1	1.4	3.8	0.2	0.3	0.4
5.0	0.5	0.8	1.0	3.5	0.1	0.2	0.3
6.0	0.8	1.1	1.1	3.6	0.2	0.3	0.3
7.0	0.7	1.1	1.4	3.9	0.2	0.3	0.4
8.0	0.7	0.9	1.4	3.5	0.2	0.3	0.4
8.8	0.9	1.2	1.3	3.9	0.2	0.3	0.3

1) Quantity of phosphate in phosphate-mixture solution of various pH-value is uniform.
2) Concentration of phosphate mixture.

Table 16

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. PO_4–C, M/5. Duration of experiment: Dec. 1–6. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
H_2O	0.5	0.9	1.1	2.4	0.2	0.4	0.5
3.0	0.9	1.2	1.3	1.9	0.5	0.6	0.7
4.0	1.2	1.6	1.7	1.6	0.8	1.0	1.0
5.0	0.4	0.6	0.8	2.5	0.2	0.2	0.3
6.0	0.6	0.7	1.0	2.2	0.3	0.3	0.5
7.0	0.6	0.7	1.0	2.1	0.3	0.3	0.5
8.0	0.6	0.9	1.2	2.4	0.3	0.4	0.5
8.8	0.8	0.9	0.9	2.6	0.3	0.3	0.3

Experiment 10

In the present experiment the concentration of phosphate mixture was M/10. The quantity of phosphate in buffer solutions of various pH-values was uniform. Results are shown in Tables 17 and 18.

Table 17

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. PO_4-C, M/10.
Duration of experiment: Feb. 22–28. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.5	0.7	1.1	3.7	0.1	0.2	0.3
4.0	0.6	0.8	1;1	3.7	0.2	0.2	0.3
5.2	0.3	0.6	1.0	4.2	0.1	0.1	0.2
5.9	0.7	1.0	1.1	3.8	0.2	0.3	0.3
6.8	0.8	0.9	0.9	3.7	0.2	0.2	0.2
7.9	0.3	0.5	0.6	3.5	0.1	0.1	0.2
8.6	0.5	0.7	1.1	3.4	0.1	0.2	0.3

Table 18

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. PO_4-C, M/10.
Duration of experiment: Feb. 22–28. 1940.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.4	0.5	0.6	2.0	0.2	0.3	0.3
4.0	0.4	0.4	0.6	2.7	0.1	0.1	0.2
5.2	0.2	0.6	0.8	2.6	0.1	0.2	0.3
5.9	0.3	0.4	0.9	2.5	0.1	0.2	0.4
6.8	0.2	0.3	0.6	1.8	0.1	0.2	0.3
7.9	0.6	0.7	0.7	2.8	0.2	0.3	0.3
8.6	0.5	c.8	c.7	2.2	0.2	0.4	0.3

Experiment II

In the present experiment the concentration of phosphate was M/25. The quantity of phosphate in buffer solutions of various pH-values was uniform. Results are shown in Tables 19 and 20.

Table 19

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. PO_4-C, M/25. Duration of experiment: Feb. 27–Mar. 4. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.1	1.2	1.4	1.6	3.3	0.4	0.4	0.5
4.0	0.9	0.9	1.3	3.5	0.3	0.3	0.4
5.1	0.9	1.1	1.7	3.6	0.3	0.3	0.5
6.0	1.2	1.3	1.5	3.3	0.4	0.4	0.5
6.8	0.6	0.6	1.0	4.0	0.2	0.2	0.3
8.0	0.6	1.0	1.3	3.9	0.2	0.3	0.3
8.5	1.0	0.9	1.1	3.7	0.3	0.2	0.3

Table 20

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. PO_4-C, M/25. Duration of experiment: Feb. 27–Mar. 4. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.1	0.8	1.0	1.2	2.2	0.4	0.5	0.5
4.0	0.4	0.5	1.1	2.0	0.2	0.3	0.5
5.1	0.4	0.5	0.9	2.5	0.2	0.2	0.4
6.0	0.6	0.8	1.5	2.3	0.3	0.3	0.7
6.8	0.6	0.6	1.2	2.1	0.3	0.3	0.6
8.0	0.7	0.8	1.3	2.2	0.3	0.4	0.6
8.5	0.6	0.7	0.9	2.9	0.2	0.2	0.3

Experiment 12

In the present experiment the concentration of phosphate mixture was M/50. The quantity of phosphate in buffer solution was uniform. Results are shown in Tables 21 and 22.

Table 21

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. PO_4–C, M/50. Duration of experiment: Jan. 28–Feb. 2. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
2.9	0.5	1.0	1.1	3.9	0.1	0.3	0.3
4.2	0.8	0.9	0.9	3.2	0.3	0.3	0.3
5.1	1.0	1.3	1.7	3.8	0.3	0.3	0.4
6.0	0.9	1.1	1.3	4.4	0.2	0.3	0.3
7.0	0.9	1.1	1.3	3.9	0.2	0.3	0.3
8.0	0.8	1.0	1.1	3.2	0.3	0.3	0.3
8.6	0.5	1.0	1.1	4.0	0.1	0.3	0.3

Table 22

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. PO_4–C, M/50. Duration of experiment: Jan. 12–17. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
2.9	0.3	0.5	0.7	2.4	0.1	0.2	0.3
4.2	0.4	0.6	0.9	2.5	0.2	0.2	0.4
5.1	1.0	1.1	1.3	2.3	0.4	0.5	0.6
6.0	0.5	0.7	0.7	2.6	0.2	0.3	0.3
7.0	0.2	0.5	0.7	2.5	0.1	0.2	0.3
8.0	0.7	0.8	1.1	2.3	0.3	0.3	0.5
8.6	0.4	0.6	0.7	2.3	0.2	0.3	0.3

From the data obtained in the experiment with *P. Glehnii* (Tables 15, 17, 19 and 21) it will be recognized that there did appear an influence of hydrogen-ion concentration of the solution on the absorption of water, though the difference of absorption in the solutions of different pH was in general not very significant. The absorption of water by seeds of *P. Glehnii* was the least at pH 3.0 or 8.6 regardless of the concentration of phosphate mixture. The absorption of water became greater as the acidity of the solution became less and it reached the maximum at pH 4.0–5.0. The maximum point of absorption of water changed in accordance with the increasing of the concentrations of phosphate mixture. That is, in the phosphate mixture of M/5 concentration, the maximum zone was pH 3.8–4.0, but in the phosphate mixture of M/25 and M/50 concentrations, the maximum point or zone was at pH 5.0. This difference of maximum points of absorption of water may depend on the concentration of salts or ions of phosphate or sodium.

The results of the experiment with *P. jezoensis* show that the tendency of the absorption of water by seeds in the solutions of different pH was generally parallel to that of the results with *P. Glehnii*. But the maximum points of the absorption of water by seeds of *P. jezoensis* differ more or less from the some of *P. Glehnii*. Loo (1931) studied the problem of absorption of ammonia and nitrate by the root of *Zea Mays* seedlings, in relation to the concentration and the actual acidity of culture solution. He stated that the absorption curve is wave-formed having more than three points of depression and maximum points of absorption of ammonia and nitrate. He explained this phenomenon by assuming that the protoplasm is a mixture of amphoteric colloids each with its own independent iso-electric point. The writer's results did not agree with Loo's. As a cause of this disagreement the difference of experimental materials and experimental methods should be taken into consideration. But a general decision of this problem must be left to further studies in the future. On the whole, the absorption of water by seeds of *P. Glehnii* and *P. jezoensis* was better in the acidic side than in the alkaline side.

Experiment 13

In the present experiment the concentration of phosphate mixture was M/5. The quantity of sodium in phosphate mixtures of various pH-values was uniform. They were prepared in the following proportions.

pH series	Composition of phosphate mixture			Phosphate mixture cc.	Water cc.	Obtained pH	
	M/5 H_3PO_4 cc.	M/5 NaH_2PO_4 cc.	M/5 Na_2HPO_4 cc.			M/5	M/50
3.0	8.0	90.0	0	100	0	3.0	3.0
4.0	1.0	99.0	0	95.2	4.8	4.0	4.0
5.0	0	97.0	3.0	86.9	13.1	5.0	5.0
6.0	0	70.0	30.0	72.5	27.5	6.0	6.0
7.0	0	24.0	76.0	53.5	46.5	6.9	6.8
8.0	0	2.0	99.0	47.4	52.6	7.7	7.6
9.0	0	0	100.0	47.2	52.8	8.9	8.4

Results are shown in Tables 23 and 24.

Table 23

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. Na–C[1], M/5. Duration of experiment: Feb. 10–15. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.8	1.6	1.6	3.8	0.2	0.4	0.4
4.0	0.7	1.1	1.4	3.6	0.2	0.3	0.4
5.1	0.8	1.4	1.8	3.7	0.2	0.4	0.5
6.0	0.8	1.3	1.5	3.7	0.2	0.4	0.4
6.9	1.3	1.5	1.9	3.2	0.4	0.5	0.6
7.7	0.6	1.4	1.6	3.5	0.2	0.4	0.5
8.9	0.5	1.7	2.1	3.7	0.1	0.5	0.6

1) Quantity of sodium in phosphate-mixture solution of various pH-value is uniform.

Table 24

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. Na–C, M/5. Duration of experiment: Jan. 12–17. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.9	0.9	1.0	3.4	0.3	0.3	0.3
4.0	0.7	0.8	1.0	2.2	0.3	0.4	0.5
5.1	0.7	0.8	1.1	2.6	0.3	0.3	0.4
6.0	0.5	0.6	0.8	2.5	0.2	0.2	0.3
6.9	0.8	0.9	1.0	2.5	0.3	0.4	0.4
7.7	0.8	0.9	1.0	2.6	0.3	0.3	0.4
8.9	1.0	1.2	1.3	2.8	0.4	0.4	0.5

Experiment 14

In the present experiment the concentration of phosphate mixture M/50. The quantity of sodium in phosphate mixture of various pH-values was uniform. Results are shown in Tables 25 and 26.

Table 25

Amount of average increased weight of the seeds of *Picea Glehnii* in different pH-value solutions. Na–C, M/50. Duration of experiment: Feb. 10–15. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.5	1.1	2.0	3.1	0.2	0.4	0.6
4.0	0.8	1.0	1.8	3.9	0.2	0.3	0.5
5.0	0.7	1.1	1.4	3.3	0.2	0.3	0.4
6.0	0.8	1.1	1.5	3.8	0.2	0.3	0.4
6.8	0.9	1.2	1.8	3.8	0.2	0	0.5
7.6	0.8	1.3	1.7	3.4	0.2	0.4	0.6
8.4	0.9	1.0	1.3	3.1	0.3	0.3	0.4

Table 26

Amount of average increased weight of the seeds of *Picea jezoensis* in different pH-value solutions. Na–C, M/50. Duration of experiment: Jan. 28–Feb. 2. 1941.

pH	Average increased weight after (mg)			Average green weight of one seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
3.0	0.4	0.7	0.9	2.3	0.2	0.3	0.4
4.0	0.5	0.7	0.8	2.2	0.2	0.3	0.4
5.0	0.5	0.7	0.8	2.3	0.2	0.3	0.3
6.0	0.7	0.8	1.1	3.1	0.2	0.3	0.4
6.8	0.3	0.6	0.7	2.3	0.1	0.3	0.3
7.6	0.6	0.7	0.8	2.5	0.2	0.3	0.3
8.4	0.7	1.1	1.1	2.2	0.3	0.5	0.5

The favourable pH-range for absorption of water by seeds of *P. Glehnii* was pH 5.1 to 6.9 in phosphate mixture of M/5 concentration, but in the phosphate mixture of M/50 concentration the pH-range was between 5.6–6.0. In both phosphate solutions of M/5 and M/50 concentration, when the pH became smaller than 7.0 the amount of water absorbed became much more and more as has been noted, and the absorption of water was the best at pH 4.0 in the solution of M/50 concentration. In the experiment using *P. jezoensis* the favourable pH-range for the absorption of water was not in agreement with the results of the experiment using *P. Glehnii.* Also the results obtained in the mixture solution, in which uniform quantities of phosphate were added, did not agree with the results obtained in the mixture solution of equal quantities of sodium-ions. It is difficult to explain the exact cause of the difference of the two results, though, in this case, the difference of phosphate quantities and of the experimental material may be taken into consideration as a probable cause.

5 Temperature

Experiment 15

Temperature plays a great rôle in the physiological processes of living organisms, and also in pure physical and chemical relations. The following

Experiment 15 was carried out to investigate the influences of temperature on the absorption of water by seeds of *P. Glehnii* and *P. jezoensis.* The experimental methods were the same as in Exp. 2. Results are shown in Tables 27 and 28.

Table 27

Amount of average increased weight of the seeds of *Picea Glehnii* as influenced by various temperature.
Duration of experiment: Dec. 12–17. 1940.

Temperature	Average increased weight after (mg)			Average green weight of seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
10°C	1.0	1.4	1.8	3.1	0.3	0.5	0.6
20°C	0.9	1.0	1.6	3.7	0.2	0.3	0.4
30°C	1.1	1.1	1.5	3.9	0.3	0.3	0.4
33°C	0.8	1.0	1.2	3.4	0.2	0.3	0.4
40°C	1.0	1.2	1.4	3.6	0.3	0.3	0.4

Table 28

Amount of average increased weight of the seeds of *Picea jezoensis* as influenced by various temperatures.
Duration of experiment: Dec. 13–18. 1940.

Temperature	Average increased weight after (mg)			Average green weight of seed (mg)	Average increased weight per 1 mg of seed after (mg)		
	2 days	4 days	6 days		2 days	4 days	6 days
10°C	0.5	0.6	1.0	1.9	0.3	0.3	0.5
20°C	0.4	0.5	0.8	1.4	0.3	0.4	0.6
30°C	0.4	0.7	1.0	1.9	0.2	0.4	0.5
33°C	0.4	0.5	0.9	1.8	0.2	0.3	0.5
40°C	1.0	1.3	1.3	1.4	0.7	0.9	0.9

Table 27 shows that the absorption of water at 40°C is less than at other temperatures, that is, at the lower temperature the absorption of water became much greater than at higher temperatures. But Table 28 shows that the absorption of water at 40°C is greater than at lower temperatures, for example, the absorption of water at 20°C was less than at 40°C. The results with seeds of *P. Glehnii* did not agree with those of *P. jezoensis* in the relations to the temperature. The absorption of water by the latter seeds increased during 6 days parallel with the increasing of temperature, but in absorption by the former seeds the tendency was the contrary. BROWN and WORLEY (1912) studied the rate of intake of water by barley grains at different temperatures and obtained a temperature coefficient of 1.8 to 1.9, and they considered that chemical processes were involved in the penetration of water through the semipermeable seed coat of the barley grain. SHULL (1920), and SHULL and SHULL, S. P. (1924), however, found another mean value of temperature coefficient. But the result of the present investigations did not show a positive temperature coefficient as reported by BROWN and others. At any rate, it may be said that the absorption of water is influenced by the surrounding temperature. However, the relations between temperature and the absorption of water can not be shown simply, because the processes of the absorption of water by living seeds at different temperatures involve complicated physical and chemical changes and are also influenced by biological conditions of the seeds.

IV Factors influencing the swelling of seed

The results of the foregoing series of Experiments 2–15 show that the absorption of water by seeds during the process of germination is affected by various factors, for example, substances dissolved in solution, hydrogen-ion concentration and temperature. These results suggest the probability of the unfavourable or favourable influences of electrolytes or non-electrolytes on the absorption of water, in consequence, on the germination of seeds. Therefore, the absorption of water by seeds is intimately related to their swelling, and, in consequence, the process of germination, swelling of seed and the absorption of water are closely connected with each other. The following Experiments 16–18 were intended to investigate the relation between the swelling of seeds of *P. Glehnii* and *P. jezoensis* and the electrolytes, non-electrolytes, hydrogen-ion concentration and temperature. The Method of procedure was as follows: The seeds are placed on the moistened filter-paper in the PETRI-dish as described in Exp. 1. (Fig. 1. a. b.). After thus prepared PETRI-dish had been kept in the thermostat of 20°C for a definite number of days the seeds were removed on another slide glass one by one. Then the largest length and largest width of every seed were measured using objective micrometer under a microscope with an objective of 8 X and an ocular of 4 X.

1 Electrolyte

KCl, NaCl, LiCl, NH_4Cl, $CaCl_2$, $MgCl_2$, $BaCl_2$, and $AlCl_3$ were used as electrolytes.

Experiment 16

The results of this experiment are shown in Tables 29 and 30.

Table 29

Average swelling of the seeds of *Picea Glehnii* in various salt solutions.

Duration of experiment: Sept. 20–Dec. 15. 1940.

Salts	Concent-ration (mol)	Direction	Initial length and width ±0.02mm	Increased length and width of the seeds after 0.02 mm			Percentage o increased length and width
				1 day	4 days	7 days	
KCl	0.1	Length Width	3.52 1.92	0.07 0.03	0.10 0.07	0.10 0.08	3 4
	0.5	Length Width	3.45 1.72	0.07 0.03	0.07 0.08	0.08 0.08	2 5
NaCl	0.1	Length Width	3.80 1.85	0.05 0.07	0.07 0.10	0.22 0.10	6 5
	0.5	Length Width	3.57 1.68	0.10 0.08	0.10 0.12	0.10 0.12	3 7
LiCl	0.1	Length Width	3.52 1.77	0.05 0.03	0.13 0.08	0.13 0.13	4 8
	0.5	Length Width	3.35 1.62	0.05 0.05	0.08 0.07	0.10 0.10	3 6
NH_4Cl	0.1	Length Width	3.57 1.70	0.07 0.08	0.08 0.13	0.17 0.15	5 9
	0.5	Length Width	3.62 1.67	0.05 0.05	0.10 0.08	0.13 0.13	4 8

Table 29 (*Continued*)

Sa ts	Concent-ration (mol)	Direction	Initial length and width ±0.02mm	Increased length and width of the seeds after 0.02 mm			Percentage of increased length and width
				1 day	4 days	7 days	
$CaCl_2$	0.1	Length Width	3.60 1.78	0.03 0.05	0.03 0.05	0.10 0.13	3 7
	0.5	Length Width	3.27 1.63	0.07 0.05	0.12 0.10	0.12 0.15	3 10
$MgCl_2$	0.1	Length Width	3.60 1.80	0.03 0.05	0.05 0.13	0.10 0.13	3 7
	0.5	Length Width	3.45 1.72	0.03 0.05	0.10 0.08	0.10 0.08	3 5
$BaCl_2$	0.1	Length Width	3.50 1.85	0.07 0	0.15 0.08	0.15 0.10	4 6
	0.5	Length Width	3.75 1.77	0.02 0	0.02 0.05	0.07 0.10	2 6
$AlCl_3$	0.1	Length Width	3.80 1.75	0.08 0.03	0.17 0.10	0.17 0.13	5 3
	0.5	Length Width	3.72 1.90	0.05 0.03	0.10 0.08	0.12 0.08	3 5

Table 30

Average swelling of the seeds of *Picea jezoensis* in various salt solutions.
Duration of experiment: Sept. 20–Dec. 15. 1940.

Salts	Concent-ration (mol)	Direction	Initial length and width ±0.02mm	Increased length and width of the seeds after 0.02 mm			Percentage of increased length and width
				1 day	4 days	7 days	
KCl	0.1	Length Width	2.67 1.57	0.05 0.10	0.07 0.10	0.08 0.12	3 7
	0.5	Length Width	2.78 1.62	0.07 0	0.08 0.07	0.12 0.08	4 5
NaCl	0.1	Length Width	2.68 1.52	0.03 0.07	0.07 0.12	0.08 0.12	3 8
	0.5	Length Width	2.57 1.47	0.07 0.08	0.10 0.08	0.10 0.12	4 8

Table 30 (*Continued*)

Salts	Concentration (mol)	Direction	Initial length and width ±0.02mm	Increased length and width of the seeds after 0.02 mm			Percentage of increased length and width
				1 day	4 days	7 days	
LiCl	0.1	Length Width	2.57 1.48	0.07 0.05	0.13 0.12	0.13 0.12	5 8
	0.5	Length Width	3.00 1.57	0.03 0.02	0.03 0.05	0.03 0.05	1 3
NH_4Cl	0.1	Length Width	2.73 1.50	0.02 0.05	0.03 0.07	0.07 0.07	3 4
	0.5	Length Width	2.70 1.45	0.05 0	0.07 0.02	0.07 0.02	2 2
$CaCl_2$	0.1	Length Width	2.72 1.55	0.05 0.02	0.12 0.05	0.12 0.08	5 6
	0.5	Length Width	2.60 1.47	0.05 0.02	0.08 0.05	0.08 0.05	3 3
$MgCl_2$	0.1	Length Width	2.60 1.63	0.08 0.05	0.08 0.05	0.10 0.05	4 3
	0.5	Length Width	2.38 1.42	0.03 0.07	0.07 0.07	0.08 0.08	4 6
$BaCl_2$	0.1	Length Width	2.87 1.53	0.05 0.03	0.07 0.07	0.08 0.08	3 6
	0.5	Length Width	2.70 1.52	0.10 0.02	0.10 0.10	0.12 0.13	4 9
$AlCl_3$	0.1	Length Width	2.78 1.52	0.03 0.02	0.07 0.07	0.08 0.08	3 5
	0.5	Length Width	3.08 1.68	0.08 0	0.10 0	0.10 0.03	3 3

It is clear from these data that the swelling of seeds on the filter paper moistened with salt solutions varied according to the kind of salts in various degrees. That is the swelling of seeds was varied in accordance with the difference of salts, though the differences in degree of swelling due to the difference of salts were small. Generally speaking, in the solutions of KCl, NaCl, LiCl, and NH_4Cl, the swelling of seeds both of *P. Glehnii* and *P. jezoensis* was larger than that in the solutions of $MgCl_2$, $BaCl_2$, and $AlCl_3$, and then, it is clear and quite important that the percentage of swelling of seed in width is larger than in the length.

2 Non-electrolyte

As was already stated before, it seems evident that the non-electrolytes such as alcohol, glycerine, saccharose and urea may play a direct or indirect rôle in the physiological function of a plant owing to special permeability, osmotic relationships or nutrient values. For example, it was already described in fore-going pages that these non-electrolytes have influenced the absorption of water by seeds. Then, the following experiment was carried out to ascertain the influences of non-electrolytes on the swelling of seeds of *P. Glehnii* and *P. jezoensis.*

Experiment 17

The results of Experiment 17, recorded in Tables 31 and 32, show that the effect of saccharose solution on the swelling of seeds of *P. Glehnii* is not clear in accordance with the difference of the concentration of saccharose, though the degree of swelling in both the length and width directions is slightly larger in the lower concentrations than in the higher ones. In glycerine solution, the swelling of the seeds is larger in the lower concentration than in the higher ones, but the degree of swelling in the urea solution of various concentrations does not vary in any definite relation with the increasing or decreasing of the concentration. Such tendencies were the same in the seed of *P. jezoensis.*

The results shown in Tables 33 and 34 show that the action of alcohol solution on the swelling of seeds of *P. Glehnii* and *P. jezoensis* was greater in the lower concentrations than in the higher concentrations. This fact may be attributable to the following reason: that the permeation of alcohol to seed grain may occur and this result in a retardation of the conspicuous swelling of the seed, because, in general, alcohol acts to shrink the plant cells or plant tissues.

Table 31

Average swelling of the seeds of *Picea Glehnii* in various non-electrolyte solutions.
Duration of experiment: Dec. 10–13. 1940

Non-electrolyte	Concentration	Direction	Initial length and width ±0.02mm	Increased length and width of the seeds after ±0.02 mm			Percentage of increased length and width
				1 day	4 days	7 days	
Saccharose	H_2O	Length Width	3.80 1.67	0.08 0	0.15 0.08	0.17 0.15	4 9
	0.2	Length Width	3.62 1.75	0.03 0.03	0.12 0.08	0.12 0.08	3 5
	0.3	Length Width	3.62 1.47	0.03 0.07	0.07 0.08	0.08 0.08	2 6
	0.4	Length Width	3.50 1.77	0.08 0.15	0.08 0.20	0.10 0.23	3 14
	0.7	Length Width	3.60 1.68	0.03 0.07	0.08 0.17	0.10 0.18	3 12
Glycerine	0.1	Length Width	3.52 1.67	0.15 0.08	0.13 0.12	0.22 0.13	6 8
	0.3	Length Width	3.92 1.77	0.05 0.20	0.05 0.23	0.08 0.27	2 15
	0.6	Length Width	3.57 1.73	0.05 0.13	0.05 0.15	0.05 0.15	1 8
	0.8	Length Width	3.67 1.88	0.07 0.10	0.08 0.10	0.12 0.12	3 6
	0.9	Length Width	3.32 1.70	0.12 0.05	0.15 0.05	0.18 0.07	5 4
	1.0	Length Width	3.67 1.77	0.13 0.03	0.13 0.10	0.13 0.12	4 7
Urea	0.1	Length Width	3.55 1.72	0.05 0.08	0.12 0.15	0.12 0.18	4 12
	0.2	Length Width	3.55 1.70	0.07 0	0.08 0.10	0.08 0.10	2 6
	0.4	Length Width	3.37 1.68	0 0.08	0.03 0.12	0.07 0.15	2 9
	0.5	Length Width	3.42 1.57	0.07 0.08	0.13 0.15	0.13 0.15	4 10

Table 32

Average swelling of the seeds of *Picea jezoensis* in various non-electrolyte solutions.
Duration of experiment: Dec. 10–30. 1940.

Non-electrolyte	Concentration	Direction	Initial length and width ±0.02 mm	Increased length and width of the seeds after ±0.02 mm			Percentage of increased length and width
				1 day	4 days	7 days	
Saccharose	H_2O	Length Width	2.70 1.45	0.03 0.13	0.08 0.18	— 0.18	— 13
	0.2	Length Width	3.28 1.55	0.02 0.15	0.05 0.15	0.05 0.15	3 15
	0.3	Length Width	2.90 1.52	0.02 0.03	0 0.12	0.03 0.10	1 7
	0.4	Length Width	3.08 1.50	0 0.18	0 0.20	0.03 0.20	1 14
	0.7	Length Width	2.75 1.68	0.18 0.05	0.18 0.05	0.18 0.08	7 5
Glycerine	0.1	Length Width	3.05 1.70	0.10 0.02	0.12 0.03	0.12 0.05	4 3
	0.3	Length Width	3.20 1.77	0.02 0.03	0.02 0.03	0.03 0.08	1 5
	0.6	Length Width	2.77 1.68	0.03 0.05	0.03 0.05	0.05 0.07	2 4
	0.8	Length Width	2.78 1.58	0.02 0.03	0.02 0.05	0.07 0.05	3 3
	0.9	Length Width	2.82 1.65	0.02 0.02	0.05 0.05	0.08 0.07	3 4
	1.0	Length Width	2.90 1.60	0.05 0.05	0.07 0.05	0.08 0.05	3 3
Urea	0.1	Length Width	2.80 1.60	0.03 0.03	0.05 0.05	0.08 0.13	3 8
	0.2	Length Width	2.82 1.55	0.07 0.02	0.10 0.07	0.08 0.07	3 4
	0.4	Length Width	2.78 1.53	0.07 0.05	0.10 0.05	0.10 0.05	4 3
	0.5	Length Width	2.58 1.58	0.08 0.02	0.10 0.07	0.12 0.08	5 5

Table 33

Average swelling of the seeds of *Picea Glehnii* in alcohol solution.
Duration of experiment: Jan. 5–14. 1941.

Concentration (%)	Direction	Initial length and width ±0.02 mm	Increased length and width of the seeds after ±0.02 mm			Percentage of increased length and width
			2 days	4 days	6 days	
40	Length Width	30.16 16.83	0.16 0.58	0.25 0.58	0.50 0.83	1 4
30	Length Width	27.83 16.08	0.33 0.33	0.33 0.58	0.33 0.66	1 4
20	Length Width	25.00 16.50	0.08 0.08	0.16 0.08	0.24 0.08	0.9 0.4
10	Length Width	26.00 35.30	0.50 0.24	0.66 0.24	0.83 0.66	3 1
5	Length Width	25.00 16.00	0.66 0.50	0.75 0.75	0.83 0.75	3 4
1	Length Width	36.40 36.41	0.91 0.75	0.91 0.83	1.00 1.00	2 2

Table 34

Average swelling of the seeds of *Picea jezoensis* in alcohol solution.
Duration of experiment: Jan. 5–14. 1941.

Concentration (%)	Direction	Initial length and width ±0.02 mm	Increased length and width of the seeds after ±0.02 mm			Percentage of increased length and width
			2 days	4 days	6 days	
40	Length Width	35.91 35.66	1.08 0.50	0.83 0.66	0.83 0.91	2 2
30	Length Width	33.75 17.00	0.75 1.03	0.83 0.83	0.83 1.16	2 6
20	Length Width	32.58 17.91	0.50 0.41	1.18 0.58	1.16 0.66	3 3
10	Length Width	33.08 17.58	1.16 1.43	1.25 1.25	1.66 1.83	0.6 1
5	Length Width	36.66 18.41	0.91 0.91	1.00 1.00	1.00 1.08	2 5
1	Length Width	33.16 17.41	0.83 0.66	1.08 0.66	1.08 0.66	3 4

3 Hydrogen-ion concentration

These experiments, 18 and 19, were carried out to study the influence of hydrogen-ion concentration upon the swelling of seeds.

Experiment 18

In Experiment 18, the amount of anions in each phosphate mixture solution of various pH-values was kept equal. The composition of phosphate mixture solution was similar to that in Experiment 9. The results of this experiment using phosphate mixture of M/5 concentration are shown in Tables 35 and 36. The swelling of seeds in the solution of pH 7.0 was more significant than in the solution of other pH-values, but in the phosphate mixture solution of M/50 concentration the swelling of seeds was larger in the solutions of pH 4.2 and 8.0 than in others. Among the results concerning *P. jezoensis*, in the solutions of phosphate mixture of M/50 concentration the swelling of seeds was larger at pH 7.0 and 8.0. However, on the whole, the difference in the influence on the swelling of seeds of *P. jezoensis* due to the different hydrogen-ion concentrations was not very significant.

Experiment 19

In Experiment 19, the phosphate mixture solutions of various pH-values were prepared to keep an equal quantity of sodium ions. The methods and compositions of the phosphate mixture were the same as described above. (See Experiment 13).

As shown in Table 37, the swelling of seeds in phosphate mixture solution of M/5 concentration of various pH-values was superior in the solution of pH 7.0, but in the phosphate mixture solution of M/50 concentration the swelling was superior in the solution of pH 4.0.

From the data shown in the above three Tables 35, 36 and 37 it may be seen that the swelling of seed in the solution of M/5 concentration was superior in the less acidic or neutral zone, but in the solution of M/50 concentration the swelling of seeds was superior in the acidic zone near pH 4.0. In this case the amount of phosphate ions in the solutions may be taken into consideration as the cause of these different results. In general, from the results of Experiments 18 and 19 it may be said that the swelling of seed in length direction is larger than in the width direction in the absolute value, but in relative value, that is, swelling percentage to the initial length of seed, the swelling is greater in the width than in the length.

Table 35

Average swelling of the seeds of *Picea Glehnii*. Quantity of phosphate in phosphate-mixture solution of various pH-values is uniform. Concentration of phosphate mixture is M/5 and M/50. Duration of experiment: Dec. 5–15. 1940.

Concentration of phosphate mixture	pH series	Direction	Initial length and width	Increased length and width of the seeds after			Percentage of increased length and width
				1 day	4 days	7 days	
H_2O	5.7	Length Width	3.67 1.88	0.08 0.05	0.08 0.10	0.08 0.10	2 5
M/5	3.0	Length Width	3.48 1.72	0.05 0.03	0.12 0.08	0.12 0.08	3 5
	4.0	Length Width	3.77 1.98	0.08 0.08	0.10 0.08	0.13 0.08	3 4
	5.0	Length Width	3.52 1.77	0.05 0.02	0.08 0.05	0.08 0.07	2 4
	6.0	Length Width	3.33 1.75	0.07 0.03	0.08 0.05	0.08 0.08	2 5
	7.0	Length Width	3.33 1.68	0.08 0.12	0.10 0.12	0.13 0.18	4 11
	8.0	Length Width	3.37 1.70	0.08 0.08	0.17 0.10	0.18 0.20	5 12
	8.8	Length Width	4.02 1.05	0.08 0.15	0.17 0.15	0.15 0.17	4 10
M/50	2.9	Length Width	3.63 1.83	0.07 0.12	0.08 0.13	0.10 0.15	3 8
	4.2	Length Width	3.45 1.68	0.05 0.12	0.07 0.12	0.12 0.17	3 10
	5.1	Length Width	3.52 1.70	0.10 0.07	0.12 0.08	0.13 0.08	4 5
	6.0	Length Width	3.62 1.75	0.05 0.03	0.07 0.05	0.08 0.12	2 7
	7.0	Length Width	3.42 1.82	0.08 0.07	0.12 0.07	0.13 0.08	4 4
	8.0	Length Width	3.52 1.68	0.10 0.08	0.12 0.12	0.13 0.12	4 7
	8.6	Length Width	3.47 1.73	0.15 0.12	0.13 0.15	0.17 0.13	5 8

Table 36

Average swelling of the seeds of *Picea jezoensis.* Quantity of phosphate in phosphate-mixture solution of various pH-values is uniform. Concentration of phosphate mixture is M/5 and M/50. Duration of experiment: Dec. 5–15. 1940.

Concentration of phosphate mixture	pH-series	Direction	Initial length and width	Increased length and width of the seeds after			Percentage of increased length and width
				1 day	4 days	7 days	
H_2O	5.7	Length Width	2.78 1.52	0.08 0.08	0.08 0.12	0.12 0.15	4 10
M/5	3.0	Length Width	2.90 1.62	0.07 0.07	0.08 0.07	0.12 0.08	4 5
	4.0	Length Width	2.75 1.58	0.05 0.03	0.05 0.03	0.05 0.07	2 4
	5.0	Length Width	2.93 1.67	0.08 0.05	0.08 0.07	0.08 0.08	3 5
	6.0	Length Width	2.53 1.50	0.03 0.03	0.07 0.03	0.07 0.08	3 5
	7.0	Length Width	2.88 1.63	0.03 0.03	0.05 0.05	0.10 0.05	3 3
	8.0	Length Width	3.02 1.67	0.07 0.03	0.10 0.05	0.10 0.05	3 3
	8.8	Length Width	3.00 1.70	0.07 0.02	0.07 0.02	0.07 0.03	2 2
M/50	2.9	Length Width	2.92 1.65	0.07 0.05	0.10 0.05	0.10 0.05	3 3
	4.2	Length Width	2.72 1.53	0.03 0.02	0.10 0.03	0.13 0.02	5 1
	5.1	Length Width	3.12 1.72	0.07 0.03	0.07 0.05	0.07 0.07	2 4
	6.0	Length Width	2.82 1.62	0.05 0.05	0.05 0.05	0.08 0.08	3 5
	7.0	Length Width	2.87 1.55	0.05 0.07	0.08 0.10	0.15 0.12	5 8
	8.0	Length Width	2.72 1.65	0.05 0.05	0.08 0.05	0.15 0.05	6 3
	8.6	Length Width	2.77 1.60	0.05 0.03	0.07 0.05	0.10 0.07	4 4

Table 37

Average swelling of the seeds of *Picea jezoensis*. Quantity of sodium in phosphate-mixture solution of various pH-value is uniform. Concentration of phosphate mixture is M/5 and M/50. Duration of experiment: Jan. 7–17. 2941.

Concentration of phosphate mixture	pH-series	Direction	Initial length and width	Increased length and width of the seeds after			Percentage of increased length and width
				1 day	4 days	7 days	
M/5	3.0	Length Width	2.83 1.78	0.03 0.03	0.03 0.08	0.03 0.10	1 6
	4.0	Length Width	2.83 1.55	0 0	0.02 0.03	0.03 0.03	1 2
	5.1	Length Width	2.78 1.58	0.03 0.03	0.07 0.05	0.07 0.08	3 5
	6.0	Length Width	2.85 1.70	0.05 0.02	0.05 0.02	0.05 0.08	2 5
	6.9	Length Width	2.67 1.58	0.05 0.05	0.05 0.07	0.07 0.10	3 6
	7.7	Length Width	2.75 1.55	0.08 0.10	0.13 0.12	0.13 0.13	5 8
	8.9	Length Width	2.83 1.50	0.02 0.05	0.02 0.08	0.03 0.08	1 5
M/50	3.0	Length Width	2.63 1.50	0.27 0.10	0.28 0.13	0.30 0.15	11 10
	4.0	Length Width	2.88 1.57	0.02 0.10	0.03 0.10	0.05 0.15	2 10
	5.0	Length Width	2.75 1.62	0.05 0	0.05 0.03	0.05 0.05	2 3
	6.0	Length Width	2.92 1.63	0.05 0.07	0.05 0.08	0.08 0.08	3 5
	6.8	Length Width	2.73 1.70	0.05 0.02	0.07 0.05	0.07 0.07	3 4
	7.6	Length Width	2.63 1.57	0.05 0.02	0.07 0.02	0.07 0.05	3 3
	8.4	Length Width	2.72 1.53	0.03 0.07	0.03 0.08	0.03 0.13	1 8

4 Temperature

Experiment 20

Experiment 20 was intended to ascertain the influence of temperature on the swelling of seeds of *P. Glehnii* and *P. jezoensis*.

Table 38 shows that the swelling of seed of *P. Glehnii* varied slightly with the variation of temperature, but this variation was not so clear as to warrant any conclusions on the relation between the temperature and the swelling of seeds. But in *P. jezoensis* the swelling of seed was larger in the 40°C temperature than in any other temperature, though the swelling may be, or may not be normal, because the temperature of 40°C is too high for performance of the normal physiological function for seed germination. In this case, also, the swelling of seeds in width is larger than in the length, and it is clear that the swelling in the lower temperature of 10°C is not so inferior to that at other temperatures. These facts may show that the some ecological relations exist between the swelling of seeds of *Picea* species and the surface temperature of forest soil in the germinating season in Hokkaido.

Table 38

Average swelling of the seeds of *Picea Glehnii* and *Picea jezoensis* after being kept at various temperatures.
Duration of experiment: Dec. 10–20. 1940.

Species	Temp.	Direction	Initial length and width	Increased length and width of the seeds after		Percentage of increased length and width
				1 day	4 days	
Picea Glehnii	10°C	Length	3.12	0.05	0.12	4
		Width	1.70	0.15	0.15	9
	20°C	Length	3.25	0.07	0.12	4
		Width	1.65	0.08	0.10	6
	30°C	Length	3.47	0.08	0.17	5
		Width	1.70	0.07	0.13	8
	40°C	Length	3.43	0.07	0.23	7
		Width	1.85	0.08	0.12	6
Picea jezoensis	10°C	Length	2.85	0.03	0.03	1
		Width	1.63	0.03	0.07	4
	20°C	Length	2.65	0.05	0.05	2
		Width	1.63	0.05	0.08	5
	30°C	Length	3.10	0.03	0.03	1
		Width	1.62	0.02	0.05	3
	40°C	Length	2.63	0.12	0.17	6
		Width	1.45	0.05	0.13	9

V. Factors influencing germination

Seeds that will withstand complete drying in the air without injury retain their viability under herbarium conditions from one to one hundred and fifty years, that is to say, the seeds kept under favourable conditions of the supply of water, and oxygen and of the temperature commence to germinate even after a long dormant period. In the germinating process of seeds the following facts have been reported by many authors[1]: that some seeds quickly germinate after washing, treatment with chemicals, scarification, freezing, thawing, artificial drying storage, and favourable alternations of temperature or also that the germination is influenced by light, hydrogen-ion concentration, water, hormon, warm-bath, sea water, physical stimulation and soil. But, at the present time, reports on the relation between the germination of seeds of both *P. Glehnii* and *P. jezoensis* and surrounding factors seem very poor from either the forestry or botanical point of view. Therefore the writer has carried out the following experiments in order to observe the above mentioned relations.

Methods of germination test.—As germination vessel non alkaline PETRI-dishes of 10 cm diameter and 1.5 cm depth with two sheets of filter papers were used. An adequate amount of water or solution of salts, electrolytes or phosphate mixture solution were poured into this PETRI-dish. After one week the solution in the PETRI-dish or the PETRI-dish itself was renewed in order to avoid the change of concentration of the solution on account of vaporization or action of micro-organisms. Furthermore, in Experiment 21 the weight of PETRI-dish with seeds, water and filter papers was measured occasionally for the retention constantly of the same amount of water in the PETRI-dish. All tests were run in triplicate. Since the seeds are small, 100 could be used in each dish, making 300 for each test. With this number the error of random sampling is reduced to a minimum. Thus prepared PETRI-dishes with seeds were incubated at 20°C in a thermostat for germination.

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1) MAZÉ (1900), BIRGER (1907), FISCHER (1907, 1935), DOBY (1909), SCHOTTE (1911), PROMSY (1912), BOKORNY (1913), SHULL (1914), RUDOLFS (1922, 1925), TOUMEY and DURLAND (1923), LUNDEGÅRDH (1924), LARSEN (1925), MORINAGA (1926), GRACANIN (1928), HAASIS (1928, 1930), MEYER (1928), BUCHINGER (1929), CHAPPUZEAU (1930), DORPH-PETERSON and MJASDRIKOWA (1931), KISSER and SCHUBERT (1934), NICHOLS (1934), PURVIS (1934), BALDWIN (1935), CHOLODNY (1935), BORRISS (1936, 1940), PEPTZOFF (1936), MORK (1937), RESÜHR (1939), RÖHMEHDER (1939).

1 Water

Experiment 21 was carried out to determie the influence of the amout of water in PETRI-dish upon the germination of the seeds.

Experiment 21

Seeds used were those of *P. Glehnii* and *P. jezoensis*. Results are shown in Tables 39 and 40 and Fig. 8 and 9.

From the results shown in Tables 39 and 40 it is evident that the percentage of germination of both seeds in distilled water more than 10 cc. at 30°C is low, namely, the percentage of germination of *P. Glehnii* in PETRI-dish with 10 cc. of water was 11 percent and that of *P. jezoensis*

Table 39

Germination record of the seeds of *Picea Glehnii*.
Duration of experiment: 1) Jan. 12–Feb. 2. 1940.
2) Feb. 2–Feb. 34. 1940.

Amount of water in medium (cc)	Number of germinated seed after											Percentage of germination
	6 days	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–22 "	Total	
15 30°C	0	0	0	0	0	0	0	0	1	2	3	3
10 "	0	0	1	0	1	1	1	1	1	5	11	11
6 "	0	0	1	3	2	2	2	2	2	7	21	21
4 "	1	2	4	3	4	5	2	3	4	5	33	33
15 20°C	0	2	4	10	8	8	6	5	5	18	66	66
12 "	0	5	5	5	10	8	11	3	7	11	65	65
9 "	0	8	5	10	15	14	12	3	5	7	79	79
6 "	1	11	8	13	11	13	7	3	2	8	77	77
5 "	8	16	18	—	17	11	3	2	2	2	79	79
4 "	10	15	19	—	12	7	5	1	0	6	75	75
3 "	6	17	18	—	13	10	5	2	2	5	78	78
2 "	2	20	15	—	16	9	5	3	2	5	77	77

Table 40

Germination record of the seeds of *Picea jezoensis*.
Duration of experiment: 1) Jan. 12–Feb. 2. 1940.
2) Feb. 2–Feb. 24. 1940.

Amount of water in medium (cc)	Number of germinated seed after											Percentage of germination
	6 days	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–22 "	Total	
15 30°C	0	0	1	0	1	0	0	0	0	1	3	3
10 "	0	0	0	3	2	1	0	1	0	0	7	7
6 "	0	2	2	5	2	4	1	3	3	8	30	30
4 "	2	3	8	4	9	2	0	3	3	8	42	42
15 20°C	0	7	5	12	12	12	7	2	4	4	65	65
12 "	0	8	9	18	13	11	3	2	1	1	66	66
9 "	1	9	15	17	13	7	5	1	0	3	71	71
6 "	1	12	16	19	10	5	4	1	2	0	70	70
5 "	0	10	18	—	22	8	3	2	1	3	67	67
4 "	2	13	16	—	13	9	4	1	1	2	61	61
3 "	0	7	9	—	17	12	4	1	1	3	54	54
2 "	0	6	13	—	15	9	5	1	0	2	51	51

was 7 percent. The germination of both seeds was adquate at 20°C regardless of the amount of water, but the percentage of germination became less in accordance with the increasing of water, though the dropping of the percentage was very slight. At any rate from these data it may be seen that the soaking of seed in an excess of water is harmful, that the influences of the amount of water upon the germination are intimately related with the temperature and that at 20°C there was not seen any marked difference in the tendency of dropping of the percentage of germination between the seeds of *P. Glehnii* and those of *P. jezoensis*.

Fig. 8

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* in the PETRI-dishes supplied with water of different amounts and different temperatures. Concentration in mol.

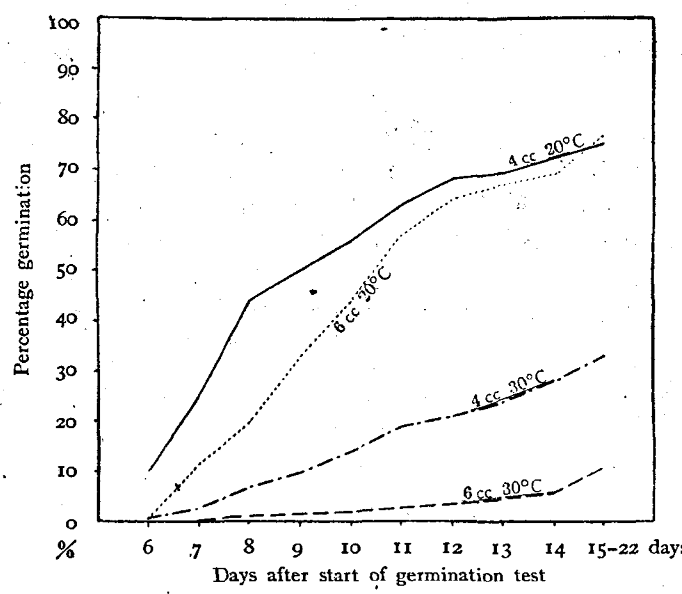
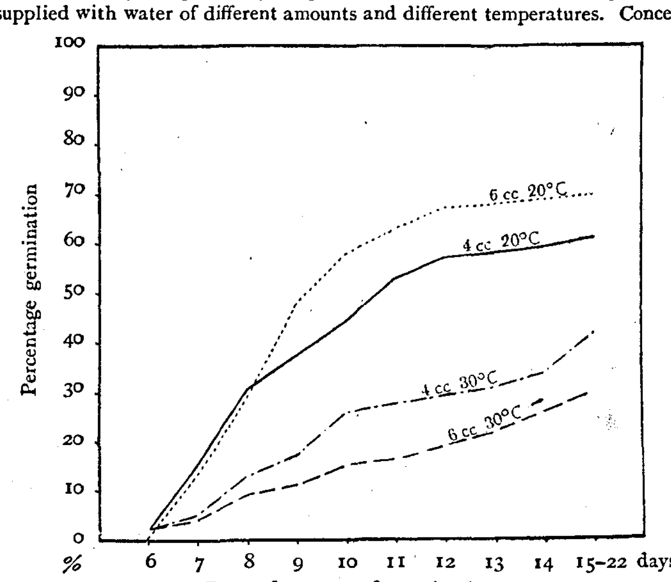


Fig. 9

Curves of daily change in percentage of germination of the seeds of *Picea jezoensis* in the PETRI-dishes supplied with water of different amounts and different temperatures. Concentration in mol.



2 Electrolyte

Experiment 22

Potassium chloride. Results are shown in Tables 41 and 42, and Fig. 10.

Table 41

Germination record of the seeds of *Picea Glehnii* in KCl-solution. Temp. 20°C.
Duration of experiment: 1) Dec. 27. 1939–Jan. 11. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	Total	
1.000 2)	0	0	0	0	0	0	0	0	0	0	0
0.500 2)	0	0	0	0	0	0	0	0	0	0	0
0.100 1)	0	4	18	15	6	2	—	8	2	55	54
0.010 1)	13	21	21	5	7	6	—	4	2	78	78
0.002 1)	6	17	21	8	11	3	—	6	2	74	75

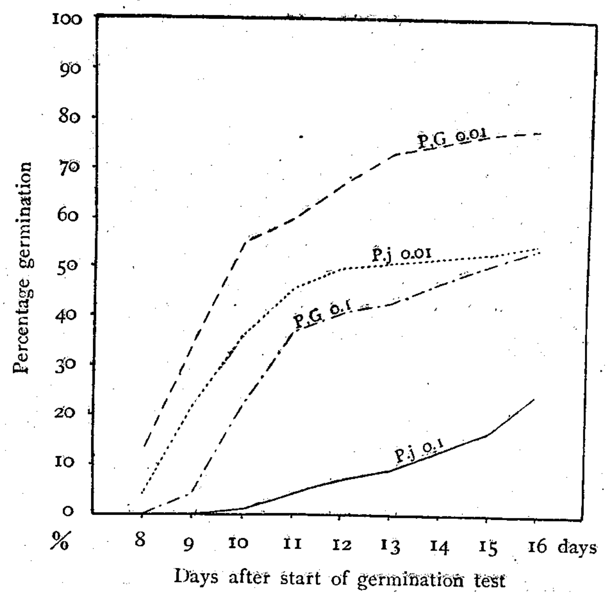
Table 42

Germination record of the seeds of *Picea jezoensis* in KCl-solution. Temp. 20°C.
Duration of experiment: 1) Dec. 27. 1939–Jan. 11. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	Total	
1.000 2)	0	0	0	0	0	0	0	0	0	0	0
0.500 2)	0	0	0	0	0	0	0	0	0	0	0
0.100 1)	0	0	1	3	3	2	—	8	8	25	26
0.010 1)	4	18	14	10	4	1	—	2	2	55	51
0.002 1)	11	17	11	6	2	1	—	1	1	50	52

Fig. 10

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in potassium chloride solution of different concentrations. Concentration in mol.



As will be seen in Table 41 the germination in KCl-solution of concentration less than 0.5 mol was good, though it decreased with the increasing of the concentration. From the previous results (Table 39) it may be said that the germination in the KCl of less than 0.01 mol concentration was normal, that is, it was nearly the same as the percentage of germination (75 %) in distilled water, but the germination in the KCl-solution of 0.1 mol concentration was 20 percent and it was less than that in the KCl-solution of more dilute concentration.

As shown in Table 42, the germination of *P. jezoensis* seeds in the KCl-solution of less than 0.01 mol concentration was very good, but that in the 0.5 mol solution was zero. That is to say, the germination decreased with the increasing of the concentration. In general, however, the germination of *P. Glehnii* seeds in the concentrated KCl-solution was less injured than that of *P. jezoensis*.

Experiment 23

Sodium chloride. Results are shown in Tables 43 and 44, and Fig. 11.

Table 43

Germination record of the seeds of *Picea Glehnii* in NaCl-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Jan. 27. 1940.
2) Feb. 37–Mar. 16. 1941.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–25 "	Total	
1.000[2]	0	0	0	0	0	0	0	0	0	0	0
0.500[2]	0	0	0	0	0	0	0	0	0	0	0
0.100[1]	2	7	13	12	10	11	—	10	4	69	69
0.010[1]	14	18	12	15	7	4	—	5	1	76	75
0.002	6	11	11	9	9	7	—	6	3	62	65

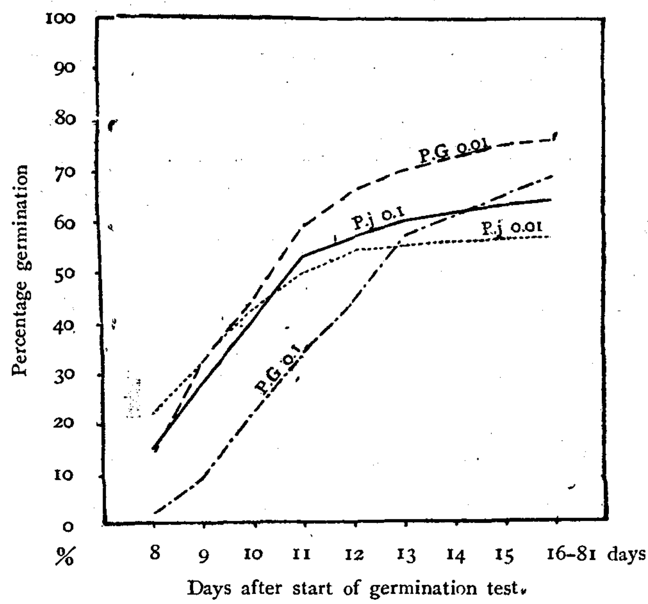
Table 44

Germination record of the seeds of *Picea jezoensis* in NaCl-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Jan. 27. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–25 "	Total	
1.000[2]	0	0	0	0	0	0	0	0	0	0	0
0.500[2]	0	0	0	0	0	0	0	0	0	0	0
0.100[1]	15	13	12	13	4	3	—	3	1	64	62
0.010[1]	22	11	10	7	3	2	—	1	0	56	57
0.002[1]	22	14	15	10	2	2	—	2	2	69	6

Fig. 11

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in sodium chloride solution of different concentrations. Concentration in mol.



The germination of *P. Glehnii* and *P. jezoensis* seeds in the NaCl-solution of 0.5 mol was zero, but it was very good in the dilute NaCl-solution of less than 0.1 mol. The germination of both seeds in NaCl-solution of the weaker concentration, was nearly the same as that in distilled water. It was concluded from the results of this experiment that both seeds would germinate as well in weaker NaCl-solution as in distilled water.

Experiment 24

Lithium chloride. Results are shown in Tables 45 and 46, and Fig. 12.

It will be seen that lithium chloride had no determinate effect upon the germination of both seeds as long as the concentration of salt remained less than 0.02 mol. That is, the seeds of *P. Glehnii* and *P. jezoensis* seem not to be extremely sensitive to lithium chloride, but *P. Glehnii* was less

Table 45

Germination record of the seeds of *Picea Glehnii* in LiCl-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Feb. 3. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–19 "	Total	
0.1000[1)]	0	0	0	0	0	0	0	0	0	0	0
0.1000[2)]	0	0	5	4	10	11	7	6	12	55	56
0.0200[2)]	0	3	7	4	9	9	3	1	3	39	42
0.0100[1)]	17	6	13	11	9	6	—	8	2	72	72
0.0100[2)]	3	7	19	11	15	9	6	3	5	78	75
0.0020[1)]	18	16	11	12	6	3	—	9	4	79	72
0.0020[2)]	6	7	19	16	8	9	3	3	4	75	75
0.0010[2)]	0	8	13	14	12	7	6	3	3	66	66
0.0001[2)]	5	13	15	12	13	10	3	2	5	78	78

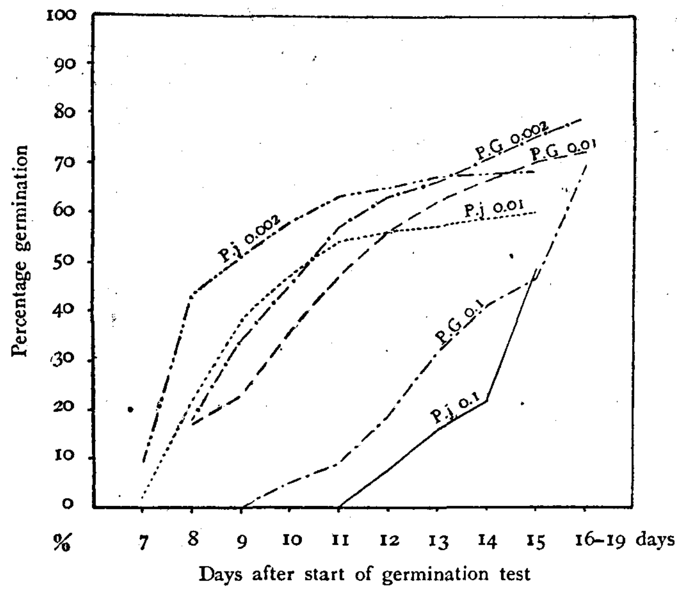
Table 46

Germination record of the seeds of *Picea jezoensis* in LiCl-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Feb. 3. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–19 "	Total	
0.1000[1)]	0	0	0	0	0	0	0	0	0	0	0
0.1000[2)]	0	0	0	0	0	0	0	0	17	39	40
0.0200[2)]	0	0	0	0	0	2	2	4	1	9	8
0.0100[1)]	2	20	16	9	7	2	1	—	3	60	59
0.0100	0	0	4	10	7	10	9	4	3	47	47
0.0020[1)]	9	34	8	7	5	2	2	—	1	68	65
0.0020[2)]	0	1	11	15	11	6	5	1	4	54	55
0.0010[2)]	0	0	2	9	7	8	7	5	8	46	43
0.0001[2)]	0	0	11	15	12	9	7	2	2	58	61

Fig. 12

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in lithium chloride solution of different concentrations. Concentration in mol.



sensitive than *P. jezoensis*. The germination of both seeds in the solution of 0.02 mol concentration was less than that in the solution of 0.1 mol or 0.01 mol concentration. No cause could be found to explain why these two kinds of seeds did not germinate better in the solution of lower concentration than in the more concentrated solution, in general, because the influence of salts upon germination may decrease with increasing of the salt concentration of solution. This salt has a tendency to change the colour of root tip to grayish, especially in the near part of root cap.

Experiment 25

Ammonium chloride. Results are shown in Tables 47 and 48, and Fig. 18.

The germination of *P. Glehnii* in a NH_4Cl-solution of 0.10 mol and of 0.01 mol was greatly less than that in distilled water, especially the percentage of germination in 0.1 mol solution was only 4 percent, but in the distilled water it was 75 percent. Also the percentage of germination in KCl-, NaCl-, and LiCl-solutions of the same concentration were nearly 65 percent; the germination in the NH_4-solution of 0.1 mol was the worst in comparison with the cases of the other salts. Considering from the above mentioned results it may be said that the less germination of *Picea* seeds in the solution of slightly higher concentration than in the lower one may be attributable to the extreme accumulation of ammonia in the seed. Though ammonium salts are absolutely necessary for the normal growth of plants, an extreme accumulation of ammonia poisins plants as described

Table 47

Germination record of the seeds of *Picea Glehnii* in NH_4Cl-solution.
Duration of experiment: Mar. 21. 1940–Apr. 12. 1940.

Concentration of salt (mol)	Number of germinated seed after											Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–22 "	Total	
1.00	0	0	0	0	0	0	0	0	0	0	0	0
0.10	0	0	0	0	0	0	1	1	0	2	4	4
0.01	1	9	6	15	8	7	2	5	1	1	55	54

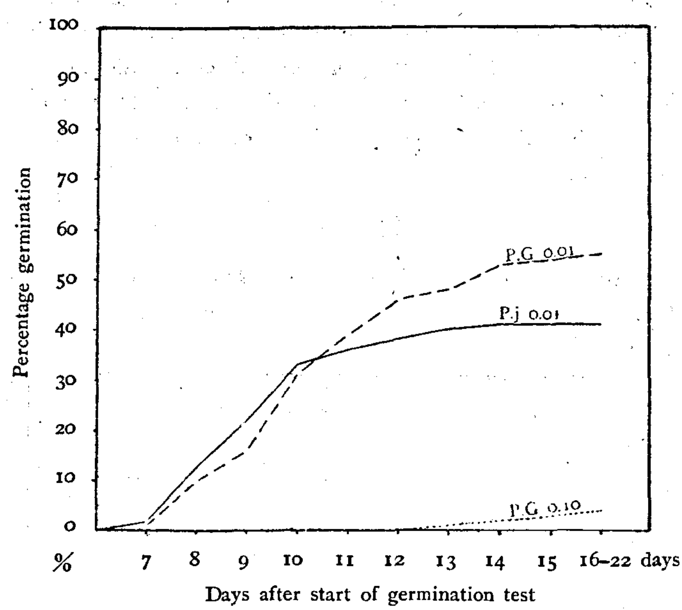
Table 48

Germination record of the seeds of *Picea jezoensis* in NH_4Cl-solution.
Duration of experiment: Mar. 21. 1940–Apr. 12. 1940.

Concentration of salt (mol)	Number of germinated seed after											Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–22 "	Total	
1.00	0	0	0	0	0	0	0	0	0	0	0	0
0.10	0	0	0	0	0	0	0	0	0	0	0	0
0.01	2	11	9	11	3	2	2	1	0	0	41	41

Fig. 13

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in ammonium chloride solution of different concentrations. Concentration in mol.



by MEVIUS (1928) and others. Of course, in this case, the deleterious effects of ammonium chloride upon the germination do not depend on the concentration itself, in other words, on the osmotic phenomenon, because the concentration of ammonium chloride of 0.1 mol is too small to cause the hypertonic action of solution.

The results obtained by using *P. jezoensis* show nearly the same tendency as in *P. Glehnii.*

Experiment 26

Calcium chloride. Results are shown in Tables 49 and 50, and Fig. 14.

The results (Tables 49 and 50) show that the retarding influence of calcium chloride solution upon germination was clear in 0.1 mol and 0.01 mol solution. That is to say, the percentage of germination of *P. Glehnii* in the 0.01 mol solution is nearly equal to that in the distilled water, but that in the 0.1 mol solution was 31 percent, which is only one half of that in the 0.01 mol solution or in distilled water. The percentage of germination of *P. jezoensis* in 0.1 mol solution was 14 percent, this value being much less than in 0.01 mol solution or in distilled water. Therefore, from the above two results it may be said that calcium chloride of higher concentration is harmful to the germination of seeds. Of course, the harmful influence may be attributable to that of ion effects of calcium not to the concentration of the salt, and then it would follow that the deleterious effects of calcium chloride upon germination were less on *P. Glehnii* than on *P. jezoensis*.

Table 49

Germination record of the seeds of *Picea Glehnii* in $CaCl_2$-solution.
Duration of experiment: 1) Jan. 10–Feb. 3. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 ”	10 ”	11 ”	12 ”	13 ”	14 ”	15 ”	16–25 ”	Total	
0.500[1)]	0	0	0	0	0	0	0	0	0	0	0
0.100[1)]	0	0	1	1	2	3	—	11	12	30	31
0.010[1)]	16	10	11	12	10	6	—	9	3	77	76
0.002[2)]	18	13	11	12	8	7	—	8	5	82	78

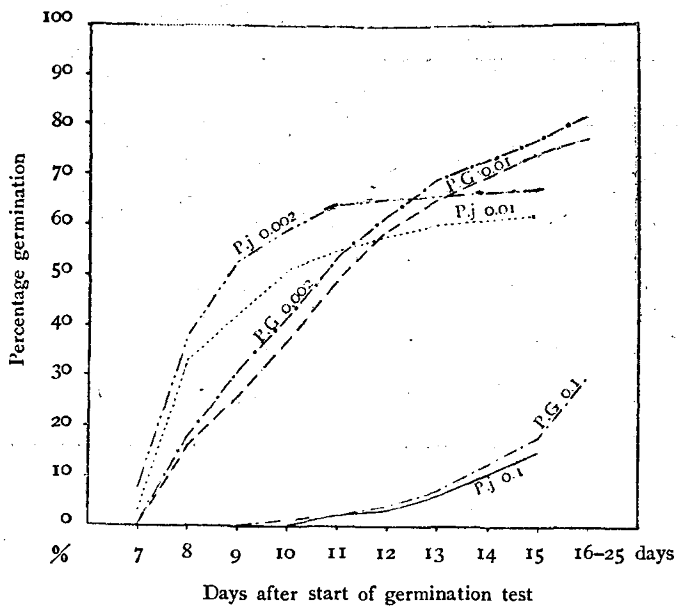
Table 50

Germination record of the seeds of *Picea jezoensis* in $CaCl_2$-solution.
Duration of experiment: 1) Jan. 10–Feb. 3. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 ”	10 ”	11 ”	12 ”	13 ”	14 ”	15 ”	16–25 ”	Total	
0.500[1)]	0	0	0	0	0	0	0	0	0	0	0
0.100[1)]	0	0	0	0	2	1	3	—	9	15	14
0.010[1)]	3	30	10	8	4	3	2	—	2	62	64
0.002[2)]	7	31	15	6	5	1	0	—	2	67	68

Fig. 14

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in calcium chloride solution of different concentrations. Concentration in mol.



Experiment 27

Magnesium chloride. Results are shown in Tables 51 and 52, and Fig. 15.

From Tables 51 and 52 it is seen that the germination of the seeds of *P. Glehnii* and *P. jezoensis* in the magnesium chloride solution of 0.1 mol was respectively 45 and 21 percent, and in the 0.01 mol or more dilute solutions they were respectively 74 and 69 percent, but in the solution of 0.5 mol solution no seed germinated in either species. Thus the relations between the concentration of $MgCl_2$ solution and germination of seeds were nearly similar to those in the case of calcium chloride, and the seeds of *P. Glehnii* were more tolerable to higher concentration of this salt than those of *P. jezoensis*.

Table 51

Germination record of the seeds of *Picea Glehnii* in $MgCl_2$-solution.
Duration of experiment: 1) Jan. 10–Jan. 17. 1940.
2) Feb. 27–Mar. 16. 1940.

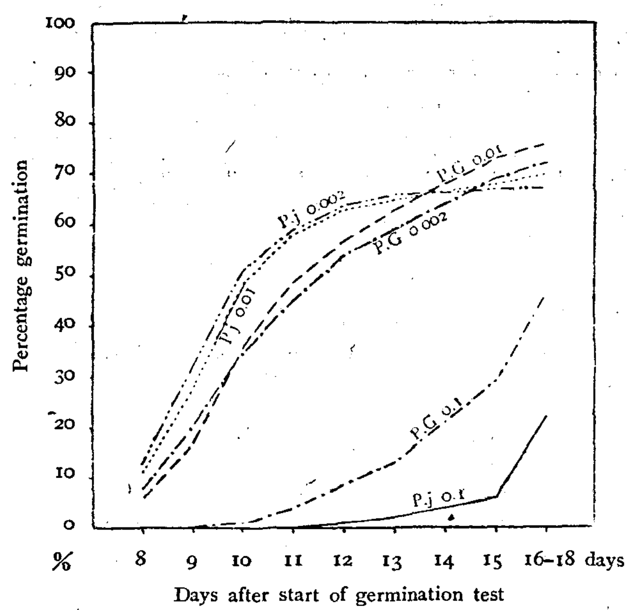
Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–18 "	Total	
0.500[2]	0	0	0	0	0	0	0	0	0	0	0
0.100[1]	0	0	1	3	5	4	—	16	17	46	45
0.010[1]	6	11	19	13	8	6	—	10	3	76	74
0.002	8	12	15	10	9	5	—	10	3	72	72

Table 52

Germination record of the seeds of *Picea jezoensis* in $MgCl_2$-solution.
Duration of experiment: 1) Jan. 10–Jan. 17. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–18 "	Total	
0.500[2]	0	0	0	0	0	0	0	0	0	0	0
0.100[1]	0	0	0	0	1	1	—	4	16	22	21
0.010[1]	11	17	20	10	5	2	—	3	2	70	69
0.002[1]	13	20	18	8	5	2	—	1	0	67	66

Fig. 15
Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in magnesium chloride solution of different concentration in mol.



Experiment 28

Barium chloride. Results are shown in Tables 53 and 54, and Fig. 16.

Table 53

Germination record of the seeds of *Picea Glehnii* in $BaCl_2$-solution. Temp. 20°C. Duration of experiment: Jan. 29–Feb. 20. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–23 "	Total	
0.100	0	0	0	0	0	0	0	0	0	0	0
0.010	5	14	15	9	13	4	4	5	8	77	78
0.002	4	17	14	17	9	6	3	3	5	78	76

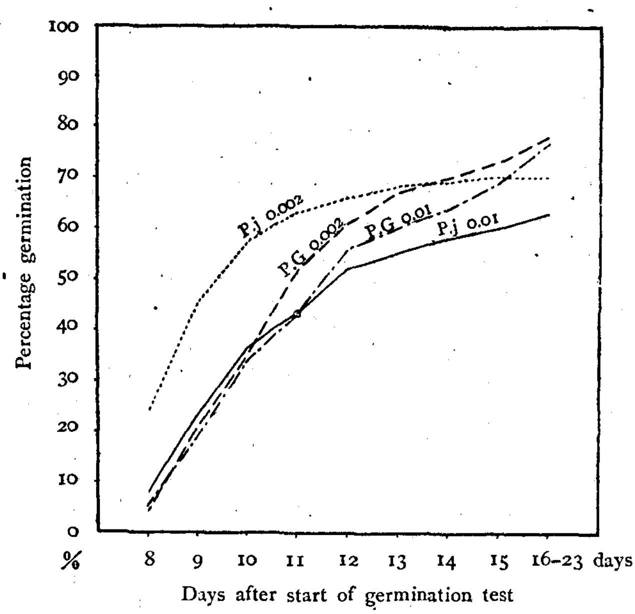
Table 54

Germination record of the seeds of *Picea jezoensis* in $BaCl_2$-solution. Temp. 20°C.
Duration of experiment: Jan. 29–Feb. 20. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–23 "	Total	
0.100	0	0	0	0	0	0	0	0	0	0	0
0.010	8	15	13	7	9	3	3	2	3	63	63
0.002	23	22	12	6	3	2	1	1	0	70	70

Fig. 16

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in barium chloride solution of different concentrations. Concentration in mol.



The results show that slightly less germination took place in the solution of 0.01 mol and the total germination of both seeds in solutions of 0.01 mol and 0.002 mol was the same as in the distilled water, but the percentage of germination of *P. Glehnii* and *P. jezoensis* in solution of 0.1 mol was zero. That is to say, 0.1 mol solutions of barium chloride in constant contact with the seeds of *P. Glehnii* and *P. jezoensis* exercised a seriously harmful influence upon their germination, as shown in Tables 53 and 54.

Experiment 29

Alminium chloride. Results are shown in Tables 55 and 56, and Fig. 17.

The germination of *P. Glehnii* seeds in the solutions of 0.0001–0.02 mol was respectively the same as that in distilled water, but it was slightly decreased in the solution of 0.1 mol, and the germination of *P. jezoensis* seeds in the solutions of various concentrations differed in each case. Such a difference in percentages of germination of *P. jezoensis* seeds according to the varying concentrations of the present salt may be attributable to the character of the seed such as imperfect fructification, because the fructification, that is seed quality, of *P. jezoensis* is inferior to that of *P. Glehnii*. But on the whole, the effects of alminium ion upon germination of *Picea* seeds were not so fatal as those of barium ion.

Table 55

Germination record of the seeds of *P. Glehnii* in $AlCl_3$-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Feb. 3. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–19 "	Total	
0.1000 1)	0	0	1	4	11	4	7	—	27	54	52
0.1000 2)	0	0	0	1	3	4	7	4	31	50	54
0.0200 2)	0	2	10	22	11	11	7	4	7	74	76
0.0100 1)	0	7	8	19	15	8	10	—	15	82	74
0.0100 2)	1	2	13	16	6	15	7	6	12	78	77
0.0020 1)	0	7	18	14	11	8	6	—	9	73	67
0.0020 2)	0	4	12	20	14	10	4	3	8	75	76
0.0010 2)	1	7	8	18	12	11	10	4	7	78	77
0.0001 2)	0	5	7	16	11	16	10	4	7	76	76

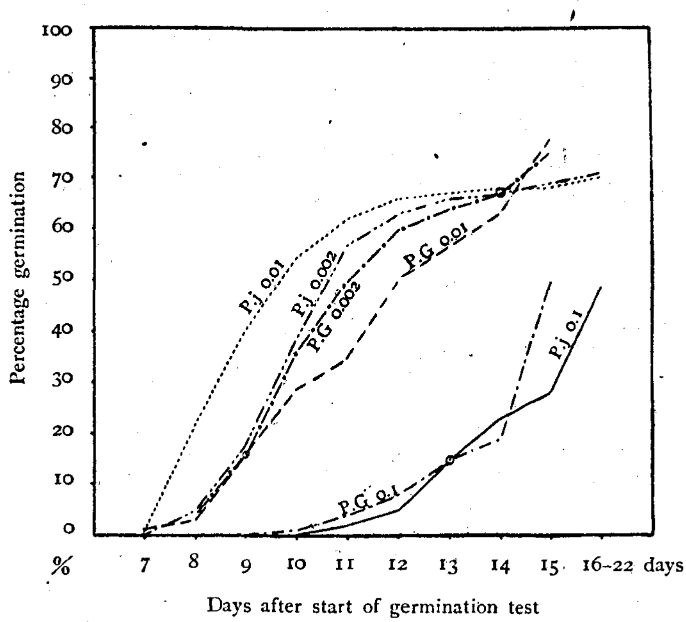
Table 56

Germination record of the seeds of *P. jezoensis* in $AlCl_3$-solution. Temp. 20°C.
Duration of experiment: 1) Jan. 10–Jan. 27. 1940.
2) Feb. 27–Mar. 16. 1940.

Concentration of salt (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–22 "	Total	
0.1000 1)	0	0	0	6	7	7	—	12	13	45	45
0.1000 1)	0	0	0	2	3	10	8	5	21	49	51
0.0200 1)	0	8	17	18	11	4	1	0	2	61	63
0.0100 2)	22	18	14	8	4	1	—	1	2	70	71
0.0100 2)	1	14	15	12	6	6	3	1	3	61	59
0.0020 2)	22	18	10	12	5	3	—	2	2	74	71
0.0020 2)	5	13	20	19	6	3	1	0	4	71	72
0.0010 2)	0	5	16	11	6	4	0	3	1	46	50
0.0010 2)	2	13	21	11	9	3	2	2	3	66	68

Fig. 17

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in alminium chloride solution in different concentrations. Concentration in mol.



3 Antagonism of salts to germination

It has already been stated before that the effect of one salt in solution upon the absorption of water or upon the swelling of seed is influenced by presence of another salt; and this phenomenon is called antagonism. From the results in the experiments described above in this paper and the review of literatures with respect to the effects of ions upon the physiological function of plants it may be considered that there also may exist some antagonistic actions between certain two kinds of salts in respect to their influences on the germination of seeds. Therefore, in order to make certain of this fact, the following Experiment 30 was performed with various combinations of several salts.

Experiment 30

The method and materials were the same as in Experiment 21. As the mixed salt solution the following combinations were prepared, using sodium chloride, potassium chloride, lithium chloride, calcium chloride, magnesium chloride and barium chloride of 0.1 mol concentration. The combinations of salt solutions are seen in Tables 55 and 56. Results are shown in Tables 57 and 58.

For the sake of simplicity, the writer has made the following summarized Tables from Tables 57 and 58.

Upon germination of *P. Glehnii* seed (a): ⟶

I series Na+K>Na+Ca>Na+Mg>Na+Ba>Na+Li (germination poor)
II series Ca+Na>Ca+Mg>Ca+K>Ca+Ba>Ca+Li
III series K+Na>K+Mg>K+Ca>K+Ba>K+Li
IV series Mg+Na>Mg+K>Mg+Ca>Mg+Ba>Mg+Li
V series Ba+Na>Ba+Ca>Ba+K>Ba+Mg>Ba+Li
VI series Li+Na>Li+Ca>Li+K>Li+Mg>Li+Ba

Upon germination of *P. jezoensis* seed (b): ⟶

I series Na+Ca>Na+K>Na+Mg>Na+Ba>Na+Li (germination poor)
II series K+Na>K+Ca>K+Mg>K+Ba>K+Li
III series Ca+Na>Ca+K>Ca+Mg>Ca+Ba>Ca+Li
IV series Mg+Na>Mg+K>Mg+Ca>Mg+Ba>Mg+Li
V series Ba+Na>Ba+Ca>Ba+K>Ba+Mg>Ba+Li
IV series Li+Ca>Li+Na>Li+K>Li+Mg>Li+Ba

Table 57

Germination record of the seeds of *Picea Glehnii*.
Duration of experiment: Apr. 20–May 13. 1940.
Concentration of salts was 0.1 mol.

Salts	Number of germinated seed after								Number of cracked seed	Percentage of germination
	9 days	10 "	11 "	12 "	13 "	14 "	15–24 "	Total		
H_2O	15	13	14	6	8	4	5	65	7	64
$CaCl_2$	0	0	0	0	1	1	23	25	44	25
Ca (9) : K (1)	0	0	1	1	3	2	18	25	39	25
Ca (8) : K (2)	0	0	0	2	2	1	26	31	38	31
Ca (4) : K (6)	0	0	0	3	7	5	10	25	43	25
KCl	0	0	0	2	2	1	20	25	33	25
K (9) : Na (1)	0	1	3	7	7	8	15	41	23	41
K (8) : Na (2)	0	1	3	6	12	9	20	51	29	50
K (4) : Na (6)	3	5	7	10	15	9	14	63	15	64
NaCl	6	13	15	13	9	3	4	63	4	63
Na (9) : Ca (1)	9	10	15	9	6	4	12	65	7	54
Na (8) : Ca (2)	0	7	11	12	14	9	15	68	13	69
Na (4) : Ca (6)	0	1	0	1	2	4	18	26	46	26
Ca (9) : Mg (1)	0	0	0	1	2	1	18	22	40	22
Ca (8) : Mg (2)	0	0	0	1	2	3	32	38	35	38
Ca (4) : Mg (6)	0	0	0	0	5	2	23	30	35	28
MgCl	0	0	0	0	0	1	10	11	21	11
Mg (9) : K (1)	0	0	1	4	6	5	24	40	29	40
Mg (8) : K (2)	0	0	1	3	3	5	24	36	37	36
Mg (4) : K (6)	0	2	0	2	2	6	25	37	47	37
Mg (9) : Na (1)	0	0	1	1	1	2	28	33	31	34
Mg (8) : Na (2)	0	1	2	3	7	4	15	32	22	32
Mg (4) : Na (6)	0	1	4	11	8	8	27	59	20	59
$BaCl_2$	0	0	0	0	0	0	0	0	23	0

Table 57 (*Continued*)

Salts	Number of germinated seed after								Number of cracked seed	Percentage of germination
	9 days	10 "	11 "	12 "	13 "	14 "	15–24 "	Total		
Ba (9) : K (1)	0	0	0	0	0	0	0	0	54	0
Ba (8) : K (2)	0	0	0	0	0	0	2	2	57	2
Ba (4) : K (6)	0	0	0	1	1	2	17	21	51	21
Ba (9) : Na (1)	0	0	0	0	0	0	0	0	57	0
Ba (8) : Na (2)	0	0	0	0	0	0	2	2	58	2
Ba (4) : Na (6)	0	0	1	3	2	4	22	32	36	33
Ba (9) : Ca (1)	0	0	0	0	0	1	8	9	48	9
Ba (8) : Ca (2)	0	0	0	0	0	0	2	2	59	2
Ba (4) : Ca (6)	0	0	0	0	0	2	28	30	43	30
Ba (9) : Mg (1)	0	0	0	0	0	0	0	0	34	0
Ba (8) : Mg (2)	0	0	0	0	0	0	0	0	27	0
Ba (4) : Mg (6)	0	0	0	1	0	0	5	6	30	6
LiCl	0	0	0	0	0	0	0	0	3	0
Li (9) : K (1)	0	0	0	0	0	0	0	0	4	0
Li (8) : K (2)	0	0	0	0	0	0	0	0	4	0
Li (4) : K (6)	0	0	0	0	1	0	4	5	18	5
Li (9) : Na (1)	0	0	0	0	0	0	0	0	6	0
Li (8) : Na (2)	0	0	0	0	0	0	0	0	7	0
Li (4) : Na (6)	0	0	1	1	0	2	8	12	21	12
Li (9) : Ca (1)	0	0	0	0	0	0	0	0	8	0
Li (8) : Ca (2)	0	0	0	0	0	0	0	0	8	0
Li (4) : Ca (6)	0	0	0	0	0	0	7	7	24	7
Li (9) : Ba (1)	0	0	0	0	0	0	0	0	6	0
Li (8) : Ba (2)	0	0	0	0	0	0	0	0	7	0
Li (4) : Ba (6)	0	0	0	0	0	0	0	0	13	0
Li (9) : Mg (1)	0	0	0	0	0	0	0	0	4	0
Li (8) : Mg (2)	0	0	0	0	0	0	0	0	6	0
Li (4) : Mg (6)	0	0	0	0	1	0	2	3	26	3

Table 58

Germination record of the seeds of *Picea jezoensis*.
Duration of experiment: Apr. 20–May 13. 1940.
Concentration of salts was 0.1 mol.

Salts	Number of germinated seed after								Number of cracked seed	Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14–24 "	Total		
H_2O	7	19	13	14	4	5	2	64	3	65
$GaCl_2$	0	0	0	0	0	0	11	11	31	11
Ca (9) : K (1)	0	0	0	0	0	1	20	21	33	21
Ca (8) : K (2)	0	0	0	2	3	7	19	31	23	31
Ca (4) : K (6)	0	0	0	1	3	5	29	38	23	38
KCl	0	0	0	3	2	3	33	41	38	41
K (9) : Na (1)	0	0	0	1	1	3	19	24	33	24
K (8) : Na (2)	0	2	3	11	15	7	17	55	7	55
K (4) : Na (6)	0	2	9	16	12	9	12	60	8	61
NaCl	1	5	9	12	8	11	16	62	8	61
Na (9) : Ca (1)	0	5	12	16	10	8	10	61	6	60
Na (8) : Ca (2)	0	2	8	12	12	9	18	61	4	60
Na (4) : Ca (6)	0	0	0	0	4	6	26	36	19	36
Ca (9) : Mg (1)	0	0	0	0	0	0	7	7	21	7
Ca (8) : Mg (2)	0	0	0	0	0	0	19	19	30	19
Ca (4) : Mg (4)	0	0	0	0	0	0	14	14	16	14
MgCl	0	0	0	0	0	0	22	22	46	22
Mg (9) : K (1)	0	0	0	0	0	1	17	18	16	18
Mg (8) : K (2)	0	0	0	0	0	1	18	19	11	20
Mg (4) : K (6)	0	0	0	1	1	2	25	29	33	29
Mg (9) : Na (1)	0	0	0	0	1	4	21	26	10	26
Mg (8) : Na (2)	0	0	0	0	1	2	27	30	26	29
Mg (4) : Na (6)	0	0	1	4	9	8	32	54	5	54
$BaCl_2$	0	0	0	0	0	0	0	0	12	0

Table 58 (*Continued*)

Salts	Number of germinated seed after								Number of cracked seed	Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14–24 "	Total		
Ba (9) : K (1)	0	0	0	0	0	0	2	2	27	2
Ba (8) : K (2)	0	0	0	0	0	0	6	6	36	6
Ba (4) : K (6)	0	0	0	0	0	1	17	18	37	18
Ba (9) : Na (1)	0	0	0	0	0	0	0	0	16	0
Ba (8) : Na (2)	0	0	0	0	0	0	3	3	40	3
Ba (4) : Na (6)	0	0	0	0	1	2	23	26	24	26
Ba (9) : Ca (1)	0	0	0	0	0	0	7	7	23	7
Ba (8) : Ca (2)	0	0	0	0	0	0	22	22	26	22
Ba (4) : Ca (6)	0	0	0	0	0	0	0	0	15	0
Ba (9) : Mg (1)	0	0	0	0	0	0	0	0	14	0
Ba (8) : Mg (2)	0	0	0	0	0	0	1	1	24	1
Ba (4) : Mg (6)	0	0	0	0	0	0	3	3	13	3
LiCl	0	0	0	0	0	0	0	0	10	0
Li (9) : K (1)	0	0	0	0	0	0	0	0	4	0
Li (8) : K (2)	0	0	0	0	0	0	0	0	6	0
Li (4) : K (6)	0	0	0	0	0	0	0	0	19	0
Li (9) : Na (1)	0	0	0	0	0	0	0	0	13	0
Li (8) : Na (2)	0	0	0	0	0	0	0	0	6	0
Li (4) : Na (6)	0	0	0	0	0	0	0	0	17	0
Li (9) : Ca (1)	0	0	0	0	0	0	0	0	7	0
Li (8) : Ca (2)	0	0	0	0	0	0	0	0	10	0
Li (4) : Ca (6)	0	0	0	0	1	1	9	11	21	11
Li (9) : Ba (1)	0	0	0	0	0	0	0	0	5	0
Li (8) : Ba (2)	0	0	0	0	0	0	0	0	6	0
Li (4) : Ba (6)	0	0	0	0	0	0	0	0	7	0
Li (9) : Mg (1)	0	0	0	0	0	0	0	0	5	0
Li (8) : Mg (2)	0	0	0	0	0	0	0	0	13	0
Li (4) : Mg (6)	0	0	0	0	0	0	0	0	19	0

As is seen in the above summarizing Tables a and b, the effects of lithium chloride and barium chloride are most harmful to compare with other salts upon the germination of *Picea* seeds. As far as shown in these two tables, HOFMEISTER's lyotropic series of ions or salts is not applicable to germination of seed, and the results shown in Tables 57 and 58 do not, also harmonize with the results of the experiments on absorption of water by seed and its swelling. That is to say, in the case of absorption of water the effects of K, Na, and Li are greater than those of Ca und Mg, and then so-called antagonistic action of ions exists between Ca and Na or K; but it does not exist between Na and K or Ca and Mg. The effect of Na and K was practically the same with respect to germination of seed, and that of Li was most unfavourable. Among bivalent cations, the action of Ba was much more powerful in the retardation of germination of seeds than other salts.

In general, the germination of *Picea* seeds was excellent generally parallel to the amount of Na or K in the mixed salt solution and it was very poor in accordance with the increasing of Ba or Li salts. So-called antagonism to the germination could not be recognized between K and L, Mg and Li, Ba and Li, Ca and Li, and Na and Li. But it may be said that the excellent effect of Na and K in a certain combination was seen in germination so far as *Picea* seeds were used, and this fact shows that the antagonistic action of ions or salts in one case does not always accord with the results in another case.

4 Non-electrolyte

The non-electrolytes differ from electrolytes or organic acids in many points such as osmotic phenomenon, ionic action, permeability, diffusion in solution or nutrient values as a nutrient source. Therefore these experiments were projected to see the relation of non-electrolytes to the germination of *P. Glehnii* and *P. jezoensis* seeds, to compare with each other the results of experiments which were performed by using electrolytes or non-electrolytes, and to see the rôle of non-electrolytes on the absorption of water by the seeds, and also the swelling and germination of the seeds. Saccharose, glycerine, urea and alcohol were used as non-electrolytes.

Saccharose

Experiment 31

Results are shown in Tables 59–62, and Figs. 18 and 19.

From the results in Tables 59–60 and Fig. 18 it may be seen that the percentage of germination of *P. Glehnii* seeds decrease in accordance with increasing of concentration of saccharose solution. The percentages of germination in solutions of less than 0.1 mol concentration was nearly the same as that in distilled water. Therefore it may be said that the saccharose solutions of less than 0.1 mol concentration did not exert harmful effects, which may depend on the osmotic action of solution, upon the germination of *P. Glehnii* and *P. jezoensis* seeds. In the solutions of more than 0.2 mol the percentages of germination of the seeds very much decreased; *P. Glehnii* seeds did not germinate any more in the solutions of more than 0.45 mol, and *P. jezoensis* seeds did not germinate in the solutions

Table 59

Germination record of the seeds of *Picea Glehnii* in saccharose solution. Temp. 20°C.
Duration of experiment: 1) Dec. 27. 1939– Jan. 11. 1940.
2) Feb. 17–Mar. 4. 1940.

Concentration of saccharose (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–18 "	Total	
1.00[1]	0	0	0	0	0	0	0	0	0	0	0
0.80[2]	0	0	0	0	0	0	0	0	0	0	0
0.60[2]	0	0	0	0	0	0	0	0	0	0	0
0.40[2]	0	0	0	0	0	0	1	0	1	2	2
0.20[2]	0	0	0	2	6	6	5	1	15	35	36
0.10[1]	2	5	21	11	13	7	—	10	1	70	70
0.02[1]	10	18	21	5	9	4	—	7	1	75	76
0.01[1]	13	17	20	6	7	3	—	5	2	73	73
H_2O[1]	11	21	19	9	3	4	—	8	0	75	74

Table 60

Germination record of the seeds of *Picea jezoensis* in saccharose solution. Temp. 20°C.
Duration of experiment: 1) Sept. 13–Oct. 5. 1940.

Concentration of saccharose (mol)	Number of germinated seed after														Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17 "	18 "	19 "	20–23 days	Total	
0.45	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0.40	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3	3
0.35	1	1	2	5	10	9	5	7	3	2	2	2	2	2	53	53

Fig. 18

Curves of daily change in percentage of germination of the seeds of *Piceà Glehnii* and in saccharose solution of different concentrations. Concentration in mol.

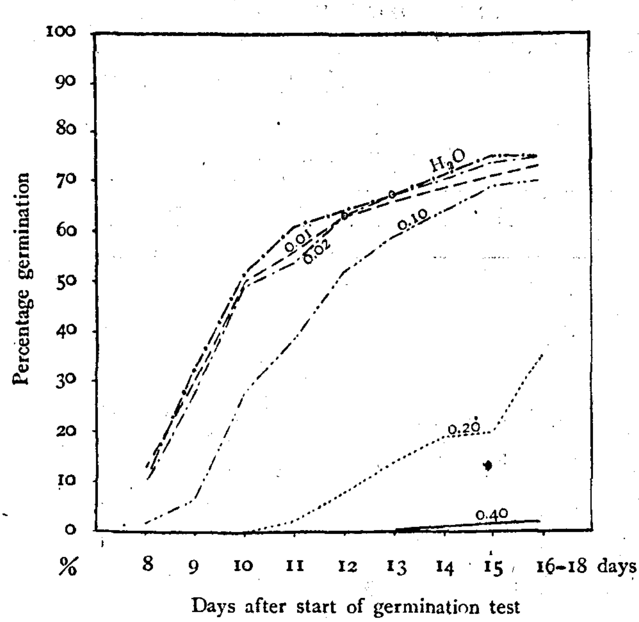


Table 61

Germination record of the seeds of *Picea jezoensis* in saccharose solution. Temp. 20°C.
Duration of experiment: 1) Dec. 27. 1939–Jan. 11. 1940.
2) Feb. 17–Mar. 4. 1940.

Concentration of saccharose (mol)	Number of germinated seed after										Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–24 "	Total	
1.00[1)]	0	0	0	0	0	0	0	0	0	0	0
0.80[2)]	0	0	0	0	0	0	0	0	0	0	0
0.60[2)]	0	0	0	0	0	0	0	0	0	0	0
0.40[2)]	0	0	0	0	0	0	0	0	0	0	0
0.20[2)]	0	0	0	0	0	0	2	1	10	13	14
0.10[1)]	0	6	14	15	10	8	—	7	1	61	61
0.02[1)]	5	19	17	13	7	3	—	2	0	66	64
0.01[1)]	8	21	16	13	5	2	—	2	1	68	69
H_2O	6	20	14	10	3	1	—	3	1	58	58

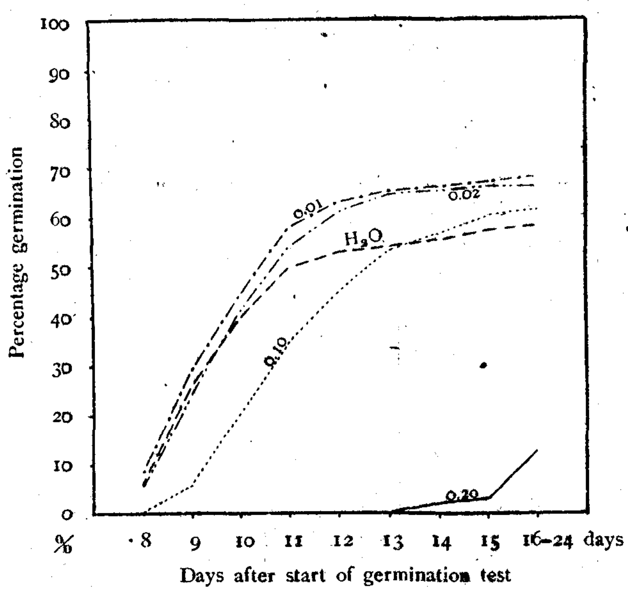
Table 62

Germination record of the seeds of *Picea jezoensis* in saccharose solution. Temp. 20°C.
Duration of Experiment: Sept. 13–Oct. 5. 1940.

Concentration of saccharose (mol)	Number of germinated seed after														Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17 "	18 "	19 "	20–23 days	Total	
0.35	0	0	0	0	0	0	2	2	3	2	2	4	4	6	25	26
0.30	0	0	0	0	0	0	0	1	0	0	0	1	1	3	6	6
0.25	2	3	7	9	15	9	4	2	2	2	1	1	0	0	57	60

Fig. 19

Curves of daily change in percentage of germination of the seeds of *Picea jezoensis* in saccharose solution of different concentrations. Concentration in mol:



higher than 0.40 mol though the former in the solution of 0.40 mol and the latter in 0.35 mol germinated moderately. They rapidly lost their capacity of germination in the solution of 0.45 mol or 0.40 mol. Judging from these facts it may be considered that the critical concentration of saccharose solution in respect to germination of *Picea* seeds may perhaps exist at 0.42 mol in *P. Glehnii* and at 0.38 mol in *P. jezoensis*. The concentration of the solution used in the present experiments throughout the capacity mol, i. e. one gram molecule of every substance in 1 L. of the solution, was adopted. In other words, in such concentration the *Picea* seeds can not germinate in the saccharose solution. The critical concentration of saccharose solution for germination of *P. Glehnii* seeds is slightly higher than that of *P. jezoensis*.

Glycerine

Experiment 32

The effect of glycerine upon germination in different concentrations was studied. Results are shown in Tables 63 and 64, and Figs 20 and 21.

Table 63 and Fig. 20 show that in concentrations of more than 1.0 mol germination of *P. Glehnii* seeds was checked, in solutions of 0.10 or 0.2 mol it was as high as in distilled water, but in solutions of 0.2–0.6 mol germination was lower than in more dilute solutions. In Table 64 and Fig. 21 it was seen that germination of *P. jezoensis* in a higher concentration than 1.0 mol was not observed, and in those solutions of less than 0.2 mol germination was very good. At any rate, germination of *P. Glehnii* and *P. jezoensis* seeds decreased in accordance with the increasing of glycerine concentration. The maximum concentration of glycerine solution

Table 63
Germination record of the seeds of *Picea Glehnii* in glycerine solution. Temp. 20°C.
Duration of experiment: 1) Sept. 13–Oct. 5. 1940.
2) Feb. 17–Mar. 5. 1940.
3) Mar. 2–Mar. 24. 1940.

Concentration of glycerine (mol)	Number of germinated seed after										Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–25 "	Total	
1.30[1]	0	0	0	0	0	0	0	0	0	0	0
1.20[1]	0	0	0	0	0	0	0	0	0	0	0
1.10[1]	0	0	0	0	0	0	0	0	0	0	0
1.00[1]	0	0	0	0	0	0	0	0	0	0	0
1.00[2]	0	0	0	0	0	0	0	0	2	2	2
0.80[3]	0	0	0	0	0	0	0	0	18	18	17
0.60[3]	0	0	0	0	1	4	11	9	30	55	53
0.40[3]	0	0	0	4	10	11	10	10	19	64	64
0.20[3]	0	3	3	6	14	9	12	9	18	74	72
0.10[2]	0	3	6	13	17	16	9	5	8	77	78
0.01[2]	3	13	15	16	13	8	4	1	4	77	77

Table 64

Germination record of the seeds of *Picea jezoensis* in glycerine solution. Temp. 20°C.
Duration of experiment: 1) Sept. 13–Oct. 5. 1940.
2) Feb. 27–Mar. 16. 1940.
3) Mar. 2–Mar. 24. 1940.

Concentration of glycerine (mol)	Number of germinated seed after										Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–25 "	Total	
1.30[1]	0	0	0	0	0	0	0	0	0	0	0
1.20[1]	0	0	0	0	0	0	0	0	0	0	0
1.10[1]	0	0	0	0	0	0	0	0	0	0	0
1.00[1]	0	0	0	0	0	0	0	0	0	0	0
1.00[2]	0	0	0	0	0	0	0	0	0	0	0
0.80[3]	0	0	0	0	0	0	0	0	4	4	4
0.60[3]	0	0	0	0	2	0	1	8	36	47	46
0.40[3]	0	0	0	3	7	10	9	15	10	54	54
0.20[3]	0	5	8	19	17	10	7	6	5	77	75
0.10[2]	0	0	13	13	19	11	3	1	5	65	64
0.01[2]	5	13	14	12	16	4	3	1	2	70	68

to allow the germination of *P. Glehnii* seeds is slightly higher than that for *P. jezoensis*. Furthermore it is interesting to note that germination of seeds of *P. Glehnii* and *P. jezoensis* does not occur in the saccharose solution of 0.4 mol notwithstanding both seeds have germinated in the glycerine solutions of 0.6 or 0.8 mol concentration. That is to say, no injury upon germination of both seeds occurred in glycerine solution higher than 0.4 mol concentration, while in dilute saccharose solution less than 0.6 mol the germination could not occur. A consideration on the cause of these different influences of saccharose and glycerine on the germination of *Picea* seeds will be presented in the later part of this paper. In Experiment 32 the hygroscopicity of glycerine must be taken into consideration in discussing the results, because as glycerine is very hygroscopic, it may be probable that the concentration of glycerine solution became more dilute in the thermostat during experiment. To clarify this point the following experiment was performed.

Fig. 20

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* in glycerine solution of different concentrations. Concentration in mol.

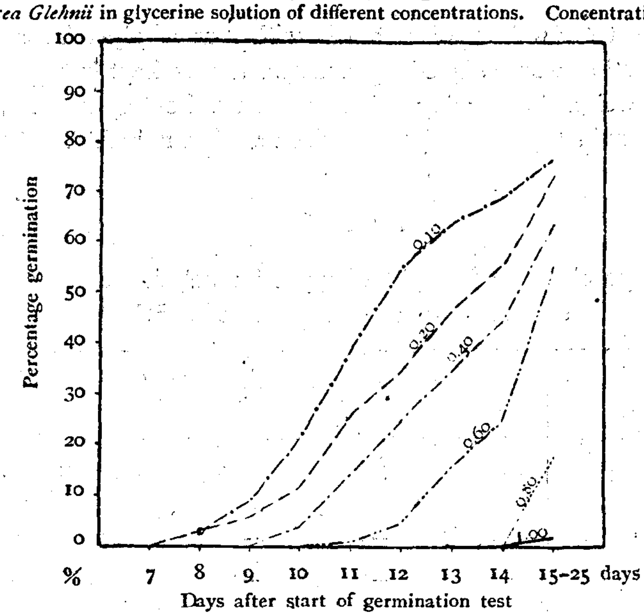
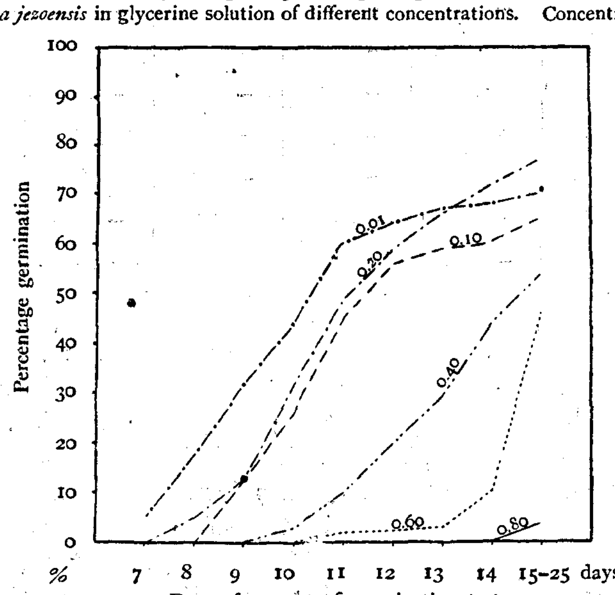


Fig. 21

Curves of daily change in percentage of germination of the seeds of *Picea jezoensis* in glycerine solution of different concentrations. Concentration in mol.



Experiment 33

After the total weight of a bottle with 5 cc of glycerine solution of each concentration was weighed by balance, it was kept in the thermostat of 20°C, and weighed every day. The bottle was kept open. Experiment was carried out in triplicate. The results are shown in Table 65.

Table 65

Average change of weight of glycerine solution of various concentration at thermostat of 20°C.

Concentration of glycerine (mol)	Initial weight	Increased or decreased weight of glycerine-solution after (grm)				Percentage of decreased weight of glycerine during 13 days (%)
		1 day	3 days	7 days	13 days	
0.05	5.0300	−0.3163	−1.5953	−3.0190	−4.8077	95
0.10	4.9912	−0.2058	−0.7097	−1.3959	−2.8603	57
0.20	5.0432	−0.2339	−0.7067	−1.4434	−2.6350	52
0.30	4.9833	−0.1952	−0.5888	−1.3788	−2.1909	43
0.40	5.0207	−0.2447	−0.8007	−1.7776	−3.1262	62
0.50	5.0481	−0.3788	−1.0575	−2.7616	−4.4034	87
0.60	5.0431	−0.3398	−0.8476	−1.7168	−3.1708	62
0.70	5.0493	−0.3162	−0.8567	−1.7409	−3.4814	68
0.80	5.1178	−0.3228	−0.7963	−1.7455	−3.6390	71
0.90	5.0925	−0.2637	−0.7551	−1.7561	−3.2983	64
1.00	5.0981	−0.2949	−0.8960	−2.0861	−3.4338	71

As is shown in the results of the present experiment the glycerine solutions of various concentration did not absorb any moisture in the thermostat, on the contrary they lost water from glycerine solution. Namely the weigt of the bottle containing glycerine solution decreased by evaporation of water, and the evaporation of water increased in accordance with the decreasing of the concentration of the solution. In general, in the germination test the decreasing of water from the germination vessel must be taken into consideration, especially in the experiment to see the relation between the concentration of salts or other substances and germination of seed, because though the concentrations of the solution were made equal at the beginning of experiment they may easily change during the experiment in thermostat.

Urea

Experiment 34

To find out the influence of urea solution of various concentrations upon germination of *Picea* seeds Experiment 34 was carried out. Results are shown in Tables 66 and 67, and Fig. 22.

In the urea solutions below 0.1 mol concentration seeds of *P. Glehnii* germinated nearly as well as in distilled water, but *P. jezoensis* seed did not germinate so well in the 0.1 mol concentration. The deleterious effect of urea solution upon germination of both seeds became serious with an increase of the concentration. The germination of the seeds of *P. Glehnii* in urea solution higher than 0.2 mol concentration was as poor as that of *P. jezoensis* seeds. More exactly, when the concentration was fixed at 0.2

Table 66

Germination record of the seeds of *Picea Glehnii* in urea solution. Temp. 20°C.
Duration of experiment: 1) Jan. 9–Feb. 16. 1940.
2) Mar. 6–Mar. 22. 1940.

Concentration of urea (mol)	Number of germinated seed after										Percentage of germination
	9 days	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–25 "	Total	
1.00[1)]	0	0	0	0	0	0	0	0	0	0	0
0.80[2)]	0	0	0	0	0	0	0	0	0	0	0
0.60[2)]	0	0	0	0	0	0	0	0	0	0	0
0.40[2)]	0	0	0	0	0	0	0	0	0	0	0
0.20[2)]	0	0	0	0	0	3	3	0	0	6	7
0.10[1)]	2	6	12	10	11	6	—	7	4	58	60
0.02[1)]	8	15	18	9	9	4	—	6	2	71	69

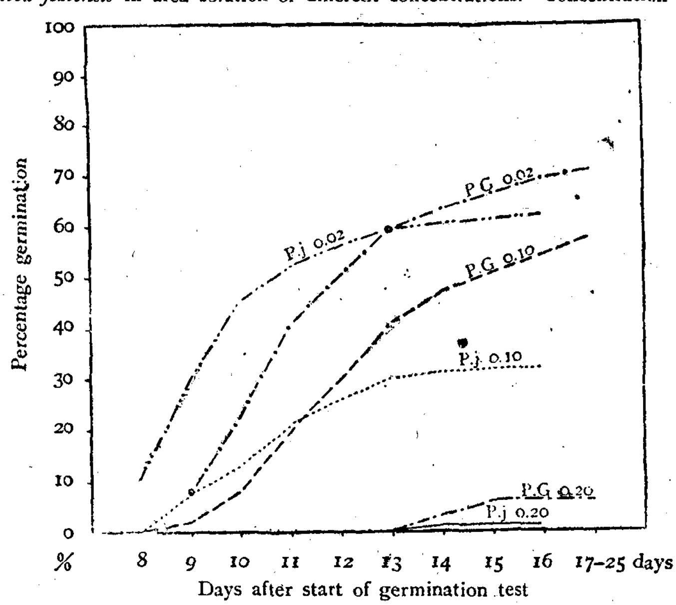
Table 67

Germination record of the seeds of *Picea jezoensis* in urea solution. Temp. 20°C.
Duration of experiment: 1) Jan. 9–Feb. 16. 1940.
2) Mar. 6–Mar. 22. 1940.

Concentration of urea (mol)	Number of germinated seed after 8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–17 "	Total	Percentage of germination
1.00[1]	0	0	0	0	0	0	0	0	0	0	0
0.80[2]	0	0	0	0	0	0	0	0	0	0	0
0.60[2]	0	0	0	0	0	0	0	0	0	0	0
0.40[2]	0	0	0	0	0	0	0	0	0	0	0
0.20[2]	0	0	0	0	0	0	1	0	0	1	1
0.10[1]	0	8	5	8	5	4	—	2	2	34	36
0.02	11	19	15	7	4	3	—	2	1	62	62

Fig. 22

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in urea solution of different concentrations. Concentration in mol.



mol the percentage of germination of *P. Glehnii* seed in saccharose solution was 36 %, in glycerine solution was 72 %, and that of *P. jezoensis* in saccharose solution was 14 %, in glycerine solution was 75 % and in urea solution was only 1 %. These results indicate that although the concentrations of these three non-electrolytes were the same, their influences upon germination of *Picea* seeds varied.

Judging from these facts it may be said that urea is very harmful upon germination of *Picea* seeds even when the concentration of urea was lower than that of saccharose or glycerine solutions. As the cause of such striking harmful effects upon germination in urea solution, the following points may be taken into consideration: firstly special permeability, secondly action of urea on physiological function of plants. In articles already published (1929, 1935), the writer has stated that the nutritive value of urea in the soil or water cultures of higher plants depends to a large extent upon the concentration of urea, the kind of plants, especially activity of urease which is contained in plant, or hydrogen-ion concentration, and he said that urea poisoning was due to extreme accumulation of urea itself and ammonia which is liberated by the action of urease from urea, to the decreasing of hydrogen-ion concentration, and to the direct effects of urea itself. Therefore, also in order to ascertain the cause of ill effects of urea upon germination one must also take into consideration the hydrogen-ion concentration, the occurrence of urease, and the detection of urea itself in *Picea* seeds submerged in urea solution. Among these points, the rôle of urease may be regarded as the most important factor, and concerning this problem the following works have been published: TAKEUCHI (1907), for the first time, demonstrated that urease exists not only in lower organisms, but also in higher plants. According to KIESEL (1927) urea can not exist unchanged in the plant body if urease occurs, and the conditions under which urease exists are closely related with the decomposition of urea. PIRSCHLE (1929) confirmed with many higher plants, especially in the root, that an intimate relation exists between the occurrence of urease and presence of urea in the culture solution. TAUBÖCK (1927), YAMAGUCHI (1930, 1935), GRANICK (1937 a, b, 1938), BOGEN (1938) also ascertained the relation between the action of urea in plant body and physiological function of plants or on the distribution of urease in plant bodies. From these investigations it may be seen that urease has a close physiological relation with the nitrogen metabolism in plant body so far as urea is produced in plant body or is resorbed as itself. Then, the following experiment was carried out in order to ascertain the above mentioned points.

Experiment 35

The seeds of *P. Glehnii* and *P. jezoensis* were germinated in PETRI-dish with distilled water in the same manner as in previous experiment. When the seeds commenced to clack or the primary root emerged, the seeds were transferred to another PETRI-dish and after sufficient washing in distilled water, 200 individual *Picia* seeds were ground in a porcelain mortar, and added with 30 cc of 30 % alcohol. Thus prepared gruel was left to stand for thirty minutes, and then filtered. Filtrates were poured into an ERLENMEYER's flask of ca 50 cc capacity with other substances.

Alcohol extract of sample	5 cc
Urea (M/100)	25 cc
Phosphate buffer mixture	5 cc
Phenol red (0.04 %)	5 drops
Thymol	a small piece

The pH-value of this mixture solution was 7.3 or 7.4 and remained nearly unchanged during the experiment. After keeping this mixture for 2 hours in the thermostat at 30°C, the total free ammonia was estimated by the micro-Kjeldahl method. As control a mixed solution was used, which was prepared using distilled water instead of 0.01 mol urea solution.

Table 68

P. Glehnii and *P. jezoensis*. 200 grains.

Seed		Seed submerged day			
		Control	2	4	6
P. Glehnii	Urea decomposed (mg) (0.03)	0.06	0.16	0.21	0.34
	Percentage of urea decomposed (%)	0.4	1.06	1.4	2.26
P. jezoensis	Urea decomposed (mg) (0.03)	0.04	0.18	0.10	0.24
	Percentage of urea decomposed (%)	0.26	1.20	0.66	1.60

Table 68 shows that the occurrence of urease in *Picea* seed was detected though its intensity varied according to the stage of germination.

Thus, from this experimental result the action of urease in germination *Picea* seeds was proved. From this fact it may be reasonable to believe that the decomposition of urea, though some of it is absorbed by seed as urea itself, occurs in germinating seed by the action of urease, consequently it results in the accumulation of ammonia and pH change. On the other hand, as the action of urease is very quick, its occurrence in the germinating seed may affect the change or transportation of urea. In order to see the change of pH-value and detect the presence of urea in germinating seeds Experiment 36 was performed.

Experiment 36

The hydrogen-ion concentration of the expressed sap of germinating seeds was determind in the following manner, using the indicator of CLARK and LUBS. A small drop of dilute indicator was put on a non-alkaline objective glass. After drying of the indicator solution, a drop of the expressed sap of the germinating seeds or very small root tip was added to it, and the pH-value was determined. The detection of urea in germinating seeds was qualitatively tested microchemically by the xanthydol method of FOSSE (1913). KLEIN and TAUBÖCK (1927) and MEVIUS (1931) pointed out that this reaction is not limited to urea, but also can be applied to ureïdes. From writer's experiment it may be reasonable to say that the crystals which appear in the tested material after the addition of the reagent indicate the reaction of urea or at least of ureïdes which can be derived from the resorbed urea.

Table 69

P. Glehnii

Concentration of urea (mol)	pH of expressed sap of germinated seed		Detection of urea
	Control (initial)	After 1 days	
0.1	4.6	8.7	−
0.2	4.6	8.9	±
0.3	4.6	9.1	+
0.4	4.6	9.3	+

From Table 69 it may be seen that the hydrogen-ion concentration of the cell sap of germinated seeds decreased extremely in accordance with the decomposition of the resorbed urea, which depends upon the amount of urea in the solution and the duration of germinating test. Furthermore, in this case there occurs an accumulation of a large amount of ammonia in germinating seeds, then it results in ammonia-poisoning as stated by MEVIUS (1928). SATO and the present writer (1939) proved that the growth of *P. Glehnii* and *P. jezoensis* seedlings in less acidic or alkaline side was very poor and in extremely alkaline side (pH 8.5) the growth was injured. Judging from these facts it may be concluded that the poor germination of *Picea* seeds in urea solution of more than 0.2 mol concentration is attributable to so-called ammonia-poisoning, to the extreme change of pH-value and to the direct influence of urea which is resorbed as urea itself by germinating seeds, but the latter factor may be most ineffectual. Because urea can not exist as urea itselff for a long time owing to the action of urease, though urea is resorbed as urea itself by seed.

Alcohol

An investigation on the influence of alcohol upon germination of *Picea* seeds was carried out in Experiment 37.

Experiment 37

Results are shown in Tables 70 and 71, and Fig. 23.

Table 70

Germination record of the seeds of *Picea Glehnii* in alcohol solution.

Duration of experiment: Mar. 21. 1940–Apr. 12. 1940.

Concentration (%)	Number of germinated seed after										Percentage of germination
	9 days	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–22 "	Total	
10	0	0	0	1	1	12	7	14	25	60	61
5	0	0	9	13	16	17	5	6	6	72	71
1	6	14	8	12	10	8	1	4	3	66	66

Table 71

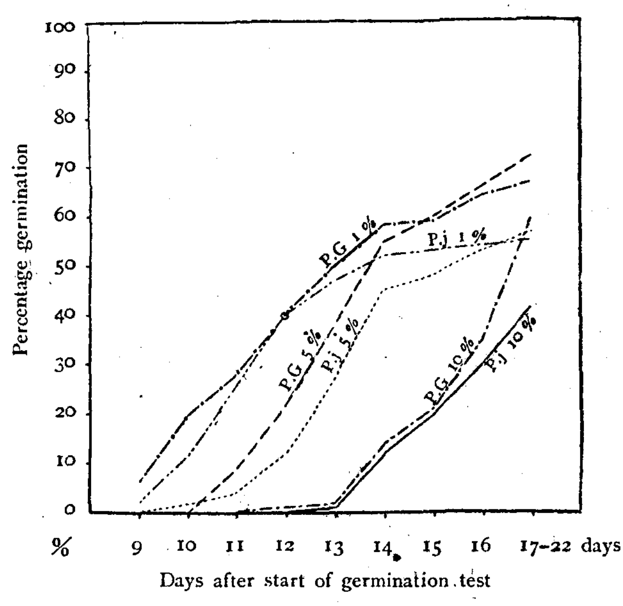
Germination record of the seeds of *Picea jezoensis* in alcohol solution.

Duration of experiment: Mar. 21. 1940–Apr. 12. 1940.

Concentration (%)	Number of germinated seed after										Percentage of germination
	9 days	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–22 "	Total	
10	0	0	0	0	1	11	8	10	12	42	42
5	0	1	3	8	15	18	3	5	4	57	58
1	2	10	4	14	7	5	1	0	2	55	54

Fig. 23

Curves of daily change in percentage of germination of the seeds of *Picea Glehnii* and *Picea jezoensis* in alcohol solution of different concentration.



The results in Tables 70 and 71 show that the germination of *Picea* seeds was influenced by concentration of alcohol, that is, the percentage of germination decreased in accordance with the increasing concentration, but the degree of decreasing of germination was not so extreme as seen in other cases such as in saccharose or urea solution. Alcohol may evaporate during experiment, because the period of germination continued 22 days in many cases. Then, in order to see the evaporation of alcohol in thermostat of various temperatures the following experiment was carried out.

Experiment 38

Twenty cc of 5 % alcohol was poured into a glass vessel of 50 cc capacity, and after keeping this alcohol solution for 1 or 2 days in the thermostat of 10°C, 20°C, 30°C, or 40°C, the total alcohol was estimated by NICLOUX's method.

Estimation of ethylalcohol.—The estimation of ethylalcohol was made according to NICLOUX's method. Given quantities (say 5 cc) of the ethylalcohol solutions of various concentrations, viz. from 0.01 to 2.00 %, were prepared, taken in a test-tube and 1 cc of 2 % potassium bichromate and 5 cc of concentrated H_2SO_4 were added, the mixture was boiled, allowed to cool. This series of test solutions showed difference in colour. By using this series as the standard, the estimation of ethylalcohol was made. Five cc of sample to be tested were put in the test-tube, and mixed with potassium bichromate and H_2SO_4 as done in the standard series, and it was compared with the corresponding standard. If the colour was intermediate between green and yellow the amount of alcohol could be at once read off.

Results are shown in Table 72.

Table 72

Concentration of alcohol 5 %.

Temperature	Alcohol (mg)		Percentage of evaporated alcohol
	Initial	Evaporated	
10°C	0.80	0.71	90%
20°C	0.80	0.75	95%
30°C	0.80	0.75	95%
40°C	0.80	0.75	95%

From this table it will be seen that the decreasing of alcohol by evaporation in the thermostat was extremely great, and that the decreasing of alcohol was greater in the thermostat of higher temperature than the lower. So that, perhaps, if the concentration of alcohol solution was kept strictly constant during experiment, the decreasing of the percentage of germination may be even larger than that shown above in Tables 70 and 71. But the decreasing of percentages of germination was clearly seen in the alcohol solution of higher concentration compared with that in the lower.

5 Hydrogen-ion concentration

The following Experiments 39–44 were carried out in order to see the influence of hydrogen-ion concentration upon the germination of the seeds of *P. Glehnii* and *P. jezoensis*. The pH series of phosphate mixture was prepared in the same manner as described in Experiment 9. In this series of experiments, the amount of phosphate in each solution with different pH-values was equal.

Experiment 39

The phosphate mixture solutions of 0.2 mol concentration were used as substrates. Results are shown in Tables 73 and 74.

Experiment 40

The concentration of the phosphate mixture solutions was 0.1 mol. Results are shown in Tables 75 ahd 76.

Experiment 41

The concentration of the phosphate mixture solution was 0.04 mol. Results are shown in Tables 77 and 78.

Experiment 42

The concentration of the phosphate mixture solution was 0.02 mol. Results are shown in Tables 79 and 80.

From Tables 73, 75, 77, 79 and Fig. 24 it may be seen that these variations in pH result in no significant difference in the rate of germination of *P. Glehnii* seeds so far as the concentration of phosphate mixture solution is less dilute than 0.04 mol, but if the concentration is 0.2 mol or 0.1 mol the influence of pH upon the germination is utterly different. In the 0.2 mol concentration the decrease in pH caused the fall of germination, in

Table 73

Germination record of the seeds of *Picea Glehnii* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was equal and the concentration of phosphate mixture was M/5. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–25 "	Total	
3.0–3.7	0	0	0	0	0	0	0	0	0	0	0	0
4.0–4.2	0	0	0	0	0	0	0	0	0	0	0	0
5.0–5.2	0	0	0	0	0	0	0	0	0	0	0	0
6.0–6.2	0	0	0	0	0	0	0	0	0	6	6	6
7.0–6.3	0	0	0	0	0	1	1	1	2	12	17	17
8.0–7.3	0	0	0	0	1	0	3	3	4	16	27	28
8.8–7.9	0	0	0	0	1	1	4	3	2	17	28	26
Control H_2O	4	3	6	4	5	2	2	2	5	16	49	48

Table 74

Germination record of the seeds of *Picea jezoensis* in the phosphate-mixture solutions with different pH-values. The amount of the sodium in each solution was equal and the concentration of phosphate mixture was M/5. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–25 "	Total	
3.0–3.7	0	0	0	0	0	0	0	0	0	1	1	1
4.0–4.2	0	0	0	0	0	0	0	0	0	2	2	2
5.0–5.2	0	0	0	0	0	0	0	0	0	1	1	1
6.0–6.3	0	0	0	0	0	0	0	3	0	5	8	8
7.0–6.4	0	0	0	0	0	0	2	5	6	13	26	26
8.0–7.8	0	0	0	0	0	2	5	4	6	18	35	34
8.8–8.2	0	0	0	0	1	1	5	6	5	8	26	26
Control H_2O	4	6	14	13	8	1	2	1	1	2	52	53

Table 75

Germination record of the seeds of *Picea Glehnii*
in the phosphate-mixture solutions with different pH-values.
The amount of phosphate in each solution was
equal and the concentration of phosphate mixture was M/10.
Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	10 days	11 "	12 "	13 "	14 "	15 "	16 "	17 "	18 "	19–21 "	Total	
3.0–3.6	3	—	13	5	5	4	2	6	3	4	45	54
4.0–4.2	1	—	4	5	5	5	2	4	1	6	33	32
5.2–5.3	0	—	5	4	3	6	2	7	1	7	35	35
5.9–5.9	1	—	3	3	4	7	0	4	2	2	26	26
6.8–6.2	3	—	9	6	5	5	2	5	1	3	39	39
7.9–7.8	4	—	14	6	1	9	2	4	1	3	44	44
8.6–8.1	4	—	8	7	5	8	1	4	4	4	45	49

Table 76

Germination record of the seeds of *Picea jezoensis*
in the phosphate-mixture solutions with different pH-values.
The amount of phosphate in each solution was
equal and the concentration of phosphate mixture was M/10.
Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	10 days	11 "	12 "	13 "	14 "	15 "	16 "	17 "	18 "	19–21 "	Total	
3.0–3.6	3	—	15	8	8	10	3	4	2	2	55	60
4.0–4.2	2	—	12	7	7	8	3	3	2	6	50	52
5.2–5.2	0	—	6	6	7	8	3	5	5	7	47	52
5.9–6.0	1	—	10	7	6	12	1	4	4	4	49	50
6.8–6.3	5	—	19	10	6	6	2	4	3	3	58	59
7.9–7.7	9	—	20	9	5	5	1	4	1	3	57	61
8.6–8.0	8	—	18	8	10	8	0	7	2	2	63	64

Table 77

Germination record of the seeds of *Picea Glehnii* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was equal and the concentration of phosphate mixture was M/25. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	6 days	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–20 "	Total	
3.1–3.2	0	1	5	12	8	9	7	6	3	4	55	57
4.0–4.2	0	1	0	12	10	13	7	6	3	6	58	59
5.1–5.1	0	1	3	6	9	8	7	10	3	7	54	55
6.0–6.1	0	1	2	6	7	8	8	4	2	6	44	47
6.8–6.4	0	1	6	7	9	8	7	5	2	4	49	49
8.0–7.8	1	2	3	11	8	10	7	3	2	5	52	52
8.5–8.1	0	2	3	12	9	9	7	5	3	4	54	53

Table 78

Germination record of the seeds of *Picea jezoensis* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was equal and the concentration of phosphate mixture was M/25. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–20 "	Total	
3.1–3.3	4	13	14	14	12	6	3	2	1	0	69	68
4.0–4.2	2	5	13	17	14	7	5	2	2	1	68	69
5.1–5.1	3	8	9	18	12	6	3	3	1	2	65	64
6.0–6.2	1	7	12	11	11	6	5	2	2	3	60	60
6.8–6.3	1	15	10	19	11	5	3	3	1	2	70	70
8.0–7.8	3	10	14	10	11	7	5	3	1	1	65	65
8.5–8.0	1	9	27	10	9	3	2	1	0	2	64	64

Table 79

Germination record of the seeds of *Picea Glehnii* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was euual and the concentration of phosphate mixture was M/50. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–20 "	Total	
2.9–3.1	0	1	4	9	7	6	6	4	5	6	48	49
4.2–4.4	0	3	10	14	9	7	4	2	4	5	58	57
5.1–5.2	0	5	7	11	9	6	6	3	3	5	55	53
6.0–6.0	1	4	11	10	7	6	5	2	2	5	53	50
7.0–6.4	0	2	7	11	9	7	7	1	3	6	53	51
8.0–7.8	0	4	10	11	7	6	7	4	1	4	54	53
8.6–8.1	1	3	7	4	6	7	6	3	1	3	41	41

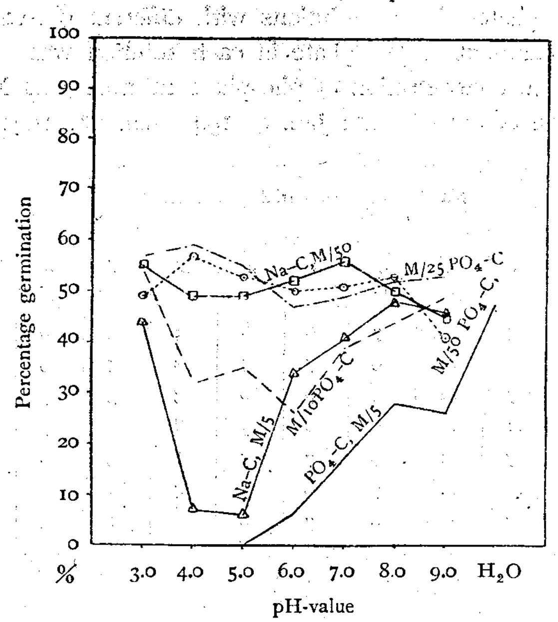
Table 80

Germinatien record of the seeds of *Picea jezoensis* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was equal and the concentration of phosphate mixture was M/50. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16–20 "	Total	
2.9–3.0	1	3	13	15	10	10	5	5	2	1	65	65
4.2–4.4	0	4	9	18	9	7	6	3	1	3	60	59
5.1–5.2	0	2	10	18	12	11	8	3	1	4	69	68
6.0–6.0	0	3	13	14	12	9	6	4	1	2	64	63
7.0–6.3	1	5	10	16	7	7	3	3	0	2	54	52
8.0–7.8	0	2	13	15	13	7	3	3	2	4	62	63
8.6–8.1	0	3	11	17	11	6	10	2	2	3	65	65

Fig. 24.

Curves of percentage of germination of the seeds of *Picea Glehnii* in solutions with various pH-values.



———— PO_4-C, M/5 (Tab. 73) ············ PO_4-C, M/50 (Tab. 79)

— — — — PO_4-C, M/10 (Tab. 75) △—△—△ Na-C, M/5 (Tab. 81)

—·—·—·— PO_4-C, M/25 (Tab. 77) □———□ Na-C, M/50 (Tab. 83)

other words the pH variations exercised great effect upon the germination. The relations among pH variation and concentration of phosphate mixture and the germination of *P. jezoensis* seeds were also the same as in that of *P. Glehnii* seeds. (Tables 74, 76, 78, 80 and Fig 24).

Accepting that as a fact could it be proved that something other than pH were the factors, which caused the retardation or harmful effects upon germination? Then, to investigate such relations more elaborately, the following experiments were undertaken using another phosphate mixture solution of various pH-value, in which the quantity of sodium was made uniform. The method of preparation of phosphate mixture solution was already described under Experiment 13.

Experiment 43

In the present experiment the concentration of the phosphate mixture solutions was 0.2 mol and the amount of sodium in each solution with different pH-values was equal. Results are shown in Tables 81 and 82.

Experiment 44

In the present experiment the concentration of the phosphate mixture solution was 0.02 mol, and the amount of sodium in each solution with different pH-values was equal. Results are shown in Tables 83 and 84, and Fig. 25.

Table 77 and 80 show that the 0.02 mol buffer solution of various pH-values had no deleterious effect on the germination of *P. Glehnii* and *P. jezoensis* seeds, but it was found that when the buffer solution of 0.2 mol concentration was used the pH variations exercised so great influence upon the germination that the percentage of germination of *P. Glehnii* and *P. jezoensis* seeds in the solution of pH 3.0 to pH 4.0 was lessened to nearly 1 % of that in the solution with pH 7.0 or more slightly on the alkaline side. In other words, if the concentration of buffer solution was more than 0.2 mol, the pH variations had great effect upon the germination of *Picea* seeds, regardless of whether the amount of phosphate or sodium in phosphate mixture solution was uniform or not.

Summarizing the above results (Tables 73–84) it may be said that the pH variations of buffer solution influenced greatly the germination of *Picea* seeds when the concentration of the buffer was great, but it was not the same when the concentration was more dilute one than 0.04 mol. What is the cause of this variation? On this point the difference of the concentration of buffer solution may be taken into consideration. Previous authors[1], who are of the opinion that the pH variations had no effect upon the germination of seeds, used a very dilute buffer solution, for example MATSUKAWA used 0.02 mol and BORTHWICK 0.0003 mol. On the other hand, FISCHER (1907), SALTER and ILVAINE (1920), ARRHENIUS (1922, 1924), LUNDEGÅRDH (1923, 1924) stated that the pH variations of solution do influence germination. They used a buffer solution of higher concentration than 0.02 mol in experiments with agricultural seeds. The writer's data obtained (Figs. 24 and 25) in the dilute concentration show that these variations in pH-value have resulted in no significant difference in percentage of germination. However, the pH

1) BORTHWICK (1936), MATSUKAWA (1936).

Table 81

Germination record of the seeds of *Picea jezoensis*
in the phosphate-mixture solutions with different pH-values.
The amount of sodium in each solution was
equal and the concentration of phosphate mixture was M/5.
Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–20 "	Total	
3.0–3.7	0	0	1	2	3	6	4	7	5	20	48	44
4.0–4.2	0	0	0	0	0	0	1	0	1	5	7	7
5.0–5.0	0	0	0	0	0	0	0	1	1	4	6	6
6.0–6.0	0	0	0	0	1	2	3	5	5	18	34	34
7.0–7.0	0	1	4	3	7	9	3	4	2	9	42	41
8.0–7.8	1	1	7	6	7	7	5	4	3	5	46	48
8.8–8.0	0	3	8	4	7	3	5	5	3	6	44	46

Table 82

Germination record of the seeds of *Picea jezoensis*
in the phosphate-mixture solutions with different pH-values.
The amount of sodium in each solution was
equal and the concentration of phosphate mixture was M/5.
Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–20 "	Total	
3.0–3.7	0	0	0	0	6	7	6	7	7	17	50	52
4.0–4.2	0	0	0	0	0	0	0	0	0	2	2	2
5.0–5.0	0	0	0	0	0	0	0	0	0	1	1	1
6.0–6.0	0	0	0	0	1	1	3	2	3	22	32	33
7.0–6.5	0	0	1	2	11	8	6	8	8	13	57	57
8.0–7.7	0	2	7	11	8	13	6	5	6	8	66	65
8.8–8.0	0	2	4	4	11	13	8	4	4	11	61	60

Table 83

Germination record of the seeds of *Picea Glehnii* in the phosphate-mixture solutions with different pH-values. The amount of sodium in each solution was equal and the concentration of phosphate mixture was M/50. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–24 "	Total	
3.0–3.6	2	10	11	8	7	5	2	5	0	4	54	55
4.0–4.1	1	8	12	5	7	3	3	3	2	2	46	49
5.0–5.1	0	5	14	7	4	4	4	3	3	4	48	49
6.0–6.0	1	6	13	8	8	7	1	2	3	2	51	52
6.8–6.4	3	12	14	7	5	3	3	3	2	4	56	56
7.6–7.4	4	9	12	5	6	6	2	2	2	2	50	50
8.4–8.0	6	7	7	5	4	4	4	3	1	3	44	45

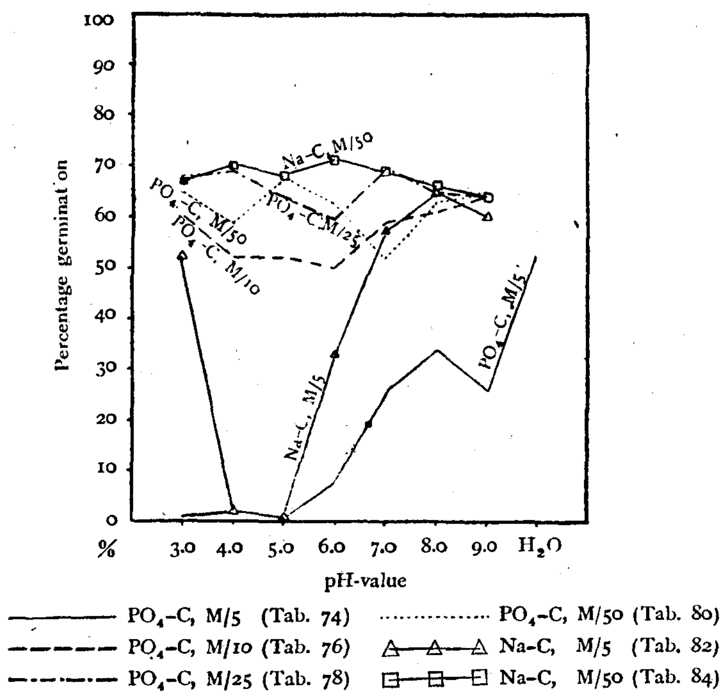
Table 84

Germination record of the seeds of *Picea jezoensis* in the phosphate-mixture solutions with different pH-values. The amount of phosphate in each solution was equal and the concentration of phosphate mixture was M/50. Duration of experiment: Jan. 9. 1941–Jan. 28. 1941.

pH	Number of germinated seed after											Percentage of germination
	8 days	9 "	10 "	11 "	12 "	13 "	14 "	15 "	16 "	17–24 "	Total	
3.0–3.7	0	2	15	13	13	8	5	5	2	3	66	67
4.0–4.2	0	8	18	14	8	8	7	2	2	3	70	70
5.0–5.0	0	6	16	11	11	11	5	3	2	3	68	68
6.0–6.0	0	5	15	17	12	11	4	2	3	2	71	71
6.8–6.5	3	9	18	12	10	7	5	3	1	1	69	69
7.6–7.2	4	12	13	12	6	12	2	1	1	2	65	66
8.4–7.6	3	9	17	12	10	5	2	3	1	2	64	64

Fig. 25

Curves of percentage of germination of the seeds of *Picea jezoensis* in solutions with various pH-values.



variations in the buffer solution of 0.2 mol concentration had a decided effect on the germination, and in an extremely small pH-value the retardation of germination was very serious. Therefore, it may be concluded that pH variations play no important rôle upon germination so far as the concentration of buffer solution was dilute, and the effect of pH variation upon germination had an intimate relation with the concentration of the solution. Thus, the concentration of buffer solution may be taken as a most responsible factor in company with pH-value in the germination of *Picea* seeds. Next, the amount of phosphate or sodium in the phosphate mixture solution varies in accordance with the shifting of the pH-value, and then in investigations on the relation between the pH-value and germination of seeds, as a matter of course, the action of sodium ions or phosphate ions on germination may be taken into consideration. However, the results obtained with the solutions of sodium constant or phosphate constant do not show remarkable differences in the effect upon germination, though the pH-value of solutions varies so far as the concentration of buffer solution is kept at less than 0.04 mol. Furthermore, also, from the results obtained in Experiment 39, in which the amount of sodium was kept uniform, and the results of Experiment 43, phosphate constant, it may be concluded that the difference of amount of sodium or phosphate can not cause the germination difference. Therefore, it may be said that the difference of sodium or phosphate ions does not cause the extreme effects upon germination. Then, in this case it is not unreasonable to consider that the pH variations of buffer solutions may be responsible for the differences of germination. That is to say, the injury to germination is due to the difference of pH-value, as can be clearly seen in Figs. 24 and 25. But the question, whether the action of pH-value is attributable to the decrease of the OH ions or the increase of H ions can not be clearly answered now, because the decreasing of pH-value, until the solution became acidic, is always accompanied by the increasing of H ions and decreasing of OH ions. But this effect of pH-values upon germination may be possible only in the case when the concentration of solution is higher than 0.04 mol, that is to say, the effect of pH-values upon germination has intimate relations with the concentration of salt in the solution.

6 Temperature

In order to study the influence of temperature upon germination of *P. Glehnii* and *P. jezoensis* seeds the following experiments were carried out.

Experiment 45

Methods and materials are the same as those described before. Results are shown in Table 85.

The results of the present experiment show clearly that the higher temperature retarded the germination of *Picea* seeds more severely than the lower temperature. The percentage of germination of *P. Glehnii* seeds at 20°C was 77 % but that at 30°C was only 26 %, and in *P. jezoensis* it was 72 % at 20°C, and 41 % at 30°C. Another experiment was carried out in the same manner as above mentioned using the seeds of *P. jezoensis* gathered in the autumn of 1939. The maturity of the seeds was very poor and the percentage of germination was very much lower comparing

Table 85

Germination record of the seeds of *Picea Glehnii* and *Picea jezoensis*.
Duration of experiment: Nov. 21–Dec. 8. 1939.

Seed	Temp.	No.	Number of germinated seed after 6 days	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14–18 days	Total	Percentage of germination
P. Glehnii	20°C	1	4	4	15	16	11	12	6	3	5	76	76
		2	3	8	30	16	13	9	3	0	4	86	78
		3	3	8	15	7	14	15	5	3	3	73	74
		Ave.	3	7	20	13	13	12	5	2	4	79	77
	30°C	1	0	0	2	3	5	3	9	4	10	36	36
		2	0	0	0	0	1	1	7	4	12	25	25
		3	0	0	2	0	2	6	2	1	5	18	18
		Ave.	0	0	1	1	3	3	6	3	9	26	26
P. jezoensis	20°C	1	2	9	19	17	9	5	2	0	3	66	66
		2	3	11	20	14	10	2	0	3	2	65	73
		3	3	19	22	13	9	6	3	1	1	77	77
		Ave.	3	13	20	15	9	4	2	1	2	69	72
	30°C	1	0	0	1	0	7	8	11	8	8	43	43
		2	0	1	2	1	6	11	8	8	5	42	42
		3	0	0	3	0	4	9	9	4	8	37	36
		Ave.	0	0	2	0	6	9	9	7	7	41	41

Table 86

Germination record of the seeds of *Picea jezoensis*.
Duration of experiment: Dec. 6–Dec. 27. 1939.

Temp.	No.	Number of germinated seed after 7 days	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–18 "	Total	Percentage of germination
20°C	1	8	1	3	1	1	0	1	1	3	19	19
	2	8	2	4	3	5	4	0	0	2	28	28
	3	9	7	3	2	1	2	0	0	0	24	24
	Ave.	8	3	3	2	2	2	0	0	1	24	24
30°C	1	3	1	0	2	0	1	2	0	4	13	13
	2	1	1	0	1	1	1	0	1	2	8	8
	3	1	1	1	0	0	0	1	1	2	7	7
	Ave.	2	1	0	1	0	1	1	1	2	9	9

with that of the seeds collected in the autumn of 1938. But the results (Table 86) obtained in the experiment, in which these poor seeds were used, also resembles the results shown in Table 85. That is, the relations between the temperature and the germination of *Picea* seeds showed exactly the same tendency in each case, though the years of gathering differed, regardless of the quality of seed.

From the results shown in Tables 39, 40, and 85, it may be said that the germination of *Picea* seeds was not good at higher temperature such as 30°C and then, in this case the amount of water plays an important rôle. Why does the slightly higher temperature exert a harmful effect upon the germination *Picea* seeds? As factors of such a harmful influence, the writer considered the change of the permeability of seed coat and the exosmosis of some substances. In order to clarify such a point the following experiment was performed.

Experiment 46

Two hundred seeds of *P. Glehnii* or *P. jezoensis* were put in a glass vessel of 50 cc capacity with 20 cc of distilled water. After these vessels were kept in the thermostat of various temperatures for a definite number of hours, the total water was brought to a definite amount by an addition of complementary volume. Then the degree of the unclearness of whitely changed solutions was determined using DUBOSQ's colorimeter. Results are shown in Table 87.

Table 87 shows that the degree of the unclearness of water, in which seeds were submerged, was greater in higher temperature than in the lower one, and that the unclearness of the solutions increased in accordance with the increasing of temperature. That is to say, the greater unclearness of solution in higher temperatures may show the greater exosmosis of substances from seeds, and in turn this greater exosmosis depends greatly on the higher permeability of seed coat. Then it may be said that the permeability of seed coat or seed itself became greater in higher temperature than in lower. But in this case, it is a question whether the greater exosmosis is due to the higher normal permeability of seed coat or to the seed itself. In general, many authors[1] have reported that the permeability of plant cells to several substances, for example KNO_3, saccharose or

1) ECKERSON (1913), STILES and JØRGENSEN (1915), STILES (1924), SEN (1928), MILLER (1931), KOSTYTSCHEW (1931).

Table 87

Temperature	Storage	No.	The ratio of clearness to water	
			P. Glehnii	*P. jezoensis*
10°C	Laboratory	1	2.83	1.98
		2	2.41	1.95
		3	2.14	1.95
	Constantly low temperature laboratory	1	2.60	2.52
		2	2.33	2.33
		3	2.10	1.95
20°C	Laboratory	1	4.59	4.10
		2	4.00	4.73
		3	4.00	3.63
	Constantly low temperature laboratory	1	6.01	8.68
		2	4.11	4.34
		3	3.25	3.41
30°C	Laboratory	1	8.22	10.40
		2	9.18	6.50
		3	8.22	8.68
	Constantly low temperature laboratory	1	6.50	7.44
		2	6.79	4.59
		3	6.50	4.89
40°C	Laboratory	1	8.22	7.44
		2	12.01	7.44
		3	8.22	12.01
	Constantly low temperature laboratory	1	14.85	8.22
		2	11.15	9.78
		3	7.80	12.01

glucose, increases gradually from a low temperature to a high temperature. Therefore it may be considered that the seed in a high temperature changes its permeability and exosmosis some substances from the seed itself and, on the other hand, these substances are available for micro-organisms as a nutrient source, and that these three factors may extremely retard the germination.

VI Absorption of non-electrolytes by the seed

It has been stated above that the non-electrolytes, such as saccharose, glycerine, urea and alcohol, have intimate relations to the absorption of water by seeds and also to their swelling and germination, and that the non-electrolytes are very different in these respects from the salts or ions thereto. In order for the non-electrolyte to exert influence on germination of seeds, it must, firstly, enter or act upon the seeds. In other words the influence of non-electrolytes on germination of seeds depends upon the permeability, or absorption of substances by the seeds. Studies on the permeability or absorption of non-electrolytes by plant cells or roots of higher plants have already been reported by many authors.[1] But the experimental reports which dealt with the problems of the absorption of non electrolytes and germination of seeds are not very satisfactory, except the following: *studies on the influence of saccharose on the respiration of* wheat and pea seeds by WOIZECHOWSKY (1903), BROWN's work (1909) on the permeability of the seeds of Hordeum vulgare, and PRINGSHEIM's (1930) ascertainment of the influence of saccharose on absorption of water by agricultural seeds.

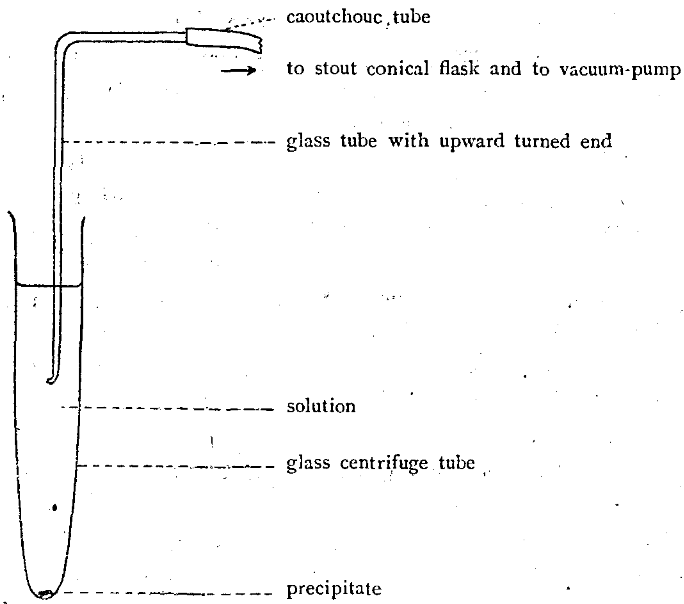
The writer carried out the following experiments to study more fully the problems concerning the influences of non-electrolytes upon the germination of seeds and, on the other hand, the absorption of non-electrolytes by *Picea* seeds. Experiments were carried out as follows; 200 individual *P. Glehnii* seeds were selected, and placed in glass vessels of 50 cc capacity with solutions of various concentrations; then they were kept in the thermostat of definite degree of temperature, the solution filtered using No. 1 Toyo filter paper, and the filtrate diluted to a definite volume using measuring flask. Then the amount of solute in filtrate was estimated by respective methods.

Estimation of saccharose.—For the estimation of saccharose, the micro-

1) KLEBS (1887), DE VRIES (1888, 1889), FITTING (1917), HÖFLER and STIEGLER (1921), HÖFLER and WEBER (1926), BÄRLUND (1929), YAMAGUCHI (1930), HÖFLER (1932, 1934, 1937), TOIVONEN (1934), BOGEN (1937, 1938), RUHLAND, ULLRICH and ENDO (1938), KREUZ (1914) and others.

BERTRAND method (KLEIN 1932) modified in some points was used combined with inversion with an aid of HCl. To the sugar solution add 4 cc of HCl of 25 %, and heat for exactly five minutes at 67–70°C. This liquid should be cooled at once, and after being neutralized with NaOH using phenolphthalein as indicator, it was diluted to a definite volume using measuring flask. (BALD, 1930). Then, put 3 cc of inverted sugar solution into one of the glass centrifuge tubes and add 3 cc of copper sulphate solution and 3 cc of alkaline tartrate solution. After this mixed solution was stirred sufficiently with glass rod, it was heated in a boiling water

Fig. 26.



bath for 6 minutes exactly, then cooled at once with running water. Next, the glass centrifuge tubes are adjusted to equal weight by adding suitable quantities of water, then centrifuged at a speed of about 2,500 revolutions per minute for 4 minutes. The precipitate should be separated from the supernant liquid and be sufficiently settled to the bottom for an easy decantation. The superntant liquid was drawn off using glass tube and vacuum-pump without disturbing the precipitates (Fig. 26). To the precipitation add about 10 cm of heated water and centrifuge again. After this process had been performed 2 times, it was dissolved by 3 cc of acid ferric sulphate solution. Then this was titrated in glass centrifuge tubes directly using ca N/60 $KMnO_4$ solution (this was prepared so that 1 cc of the solution was equivalent to 1 mg of copper).

This method described above seems to be preferable for the estimation of saccharose. In the case when the amount of saccharose corresponds to 0.5-0.10 mg of glucose, the writer has already studied the present method of the estimation of invert sugar comparing with HAGEDORN-JENSEN's method, micro-BERTRAND method, LEHMANN-MAQUENNE-SCHOORL and BERTRAND's method and prove its suitableness (not yet published). In the present case, however, the amount of saccharose in the solution is less than 0.1 mg, still this method was proved to be applicable by obtaining experimental value by analyzing saccharose solution of known concentration. HAGEDORN-JENSEN's method is the most suitable for estimation of a very small quantity of saccharose, but it is complicated and difficult in practice.

Amount of reducing sugar which correspond to the amount of copper between 0.55-17.80 mg were obtained from the Table (KLEIN 1932, p. 786), and amount of copper less than 0.55 mg is to be seen in Table 88 which was obtained from the writer's results.

Table 88

Showing amount of copper precipitated by different amounts of reducing sugars.

Glucose in milligram	Copper in milligram
0.001	0.17
0.003	0.19
0.006	0.22
0.008	0.23
0.010	0.24
0.020	0.30
0.040	0.38
0.060	0.44
0.080	0.50
0.100	0.55

Estimation of glycerine.—For the estimation of glycerine the bichromate method (MEYER 1933) was used. To 20 cc of glycerine solution of dilute concentration 25 cc of bichromate solution (74.564 g potassium bichromate and 150 cc of sulphuric acid in 1000 cc water) and 50 cc of sulphuric acid (S. G. 1. 2307) was added and after the mixture was heated on the boiling water-bath for 2 hours, it was diluted to 200 cc using measuring flask, and titrated with N/100 thiosulphate potassium iodide and starch solution. (1 cc bichromate=0.01 g glycerine). The results were calculated as shown in Table 89.

Table 89

Showing amount of glycerine as calculated by different amount of sodium thiosulphate.

N/100 $Na_2S_2O_3$ in ccm.	0	1	2	3	4	5	6	7	8	9
0	mg 0	0.065	0.130	0.195	0.260	0.325	0.390	0.455	0.520	0.585
1	0.650	0.715	0.780	0.845	0.910	0.975	1.040	1.105	1.170	1.235
2	1.300	1.365	1.430	1.495	1.560	1.625	1.690	1.755	1.820	1.885
3	1.950	2.015	2.080	2.145	2.210	2.275	2.340	2.405	2.470	2.535
4	2.600	2.665	2.730	2.795	2.860	2.925	2.990	3.055	3.120	3.185
5	3.250	3.315	3.380	3.445	3.510	3.575	3.640	3.705	3.770	3.835
6	3.900	3.965	4.030	4.095	4.160	4.225	4.290	4.355	4.420	4.485
7	4.550	4.615	4.680	4.745	4.810	4.875	4.940	5.005	5.070	5.135
8	5.200	5.265	5.330	5.395	5.460	5.525	5.590	5.655	3.720	5.785
9	5.850	5.915	5.980	6.045	6.110	6.175	6.240	6.305	6.370	6.435
10	6.500	6.565	6.630	6.695	6.760	6.825	6.890	6.955	7.020	7.085

Estimation of urea.—For the estimation of urea, the micro-Kjeldahl method for ammonia was used, combined with hydrolysis with the aid of urease. Free ammonia which is produced in consequence of decomposition of urea by the action of urease was distilled and collected by hydrochloric acid (N/100), the excess of the hydrochloric acid was titrated with sodium hydroxide (N/100), using sodium alizarin sulphonic acid as indicator. For the amount of ammonia that of urea was calculated.

Urease extract was prepared from soy bean. In the urease extract thus prepared a small quantity of free ammonia was contained. Therefore, the amount of free ammonia in the urease extract was estimated, whenever it was used, and subtracted from the amount of total ammonia in this mixture. The free ammonia produced by decomposition of urea was determined in this manner, viz., 4–5 cc of urease extract, 0.5–1.0 of phosphate buffer solution and five drops of a phenol red solution were mixed with a measured amount of sample and this mixture was placed in a thermostat of 30°C for 60–120 minutes. The pH-value of this mixed solution was nearly 7.3, which is approximately the optimum pH for the urease action (VAN SLYKE, 1914). After perfect decomposition of urea, this mixed solution was diluted properly and a certain amount was taken and distilled using the micro-Kjeldahl apparatus.

1 Saccharose

Experiment 47

After *Picea* seeds were kept in the saccharose solution of various concentrations for definite hours in the thermostat of 20°C, absorption of saccharose was estimated by analysing the solution by the above described method. Results are shown in Table 90.

Table 90

Absorption of saccharose by the seeds of *Picea Glehnii* in various concentrations of saccharose.
Figures in Table are average of 3 series.
Duration of experiment: Mar. 10–12, 1941. Temperature: 20°C.

Concentration of saccharose (mol)	Weight of seed		Absorption of saccharose (mg)	
	200 seeds (grm)	1 seed (mg)	Per 200 seeds	Per 1 seed
0.6	0.62	3.1	29.24	0.14
0.3	0.63	3.1	25.31	0.13
0.2	0.61	3.0	17.35	0.09
0.1	0.62	3.1	13.62	0.07
0.05	0.61	3.1	8.39	0.04

The data of Table 90 show that the absorption of saccharose in the higher concentration by seeds was superior to that in the lower concentration. That is to say, the higher the concentration of saccharose, the larger the absorption of saccharose was. In the 0.1 mol or 0.05 mol saccharose solution, the absorption of saccharose per seed was nearly 0.04 or 0.07 mg. This amount is very small to compare with the results shown in Table 7 and 8 (Experiment 5). From this result it may be said that the increased weight of seed, which was laid on the filter paper saturated with saccharose solution of various concentrations, depends utterly on the absorption of water, but not of saccharose.

Experiment 48

In order to ascertain the relation between the absorption of saccharose by seeds and the temperature and to consider the influence of temperature upon the absorption or germination of the seeds the present experiment was carried out. The concentration of saccharose solution was 0.1 mol. Results are shown in Table 91.

Table 91

Absorption of saccharose by the seeds of *Picea Glehnii* at various temperatures.
Duration of experiment: Jan. 11–Jan. 14. 0941.

Temp.	No.	Weight of seed		Absorption of saccharose (mg)	
		200 seeds	1 seed Average (mg)	Per 200 seeds	Per 1 seed
10°C	1	0.595	3.0	3.064	0.015
	2	0.602	3.0	2.797	0.014
	3	0.585	2.9	1.732	0.009
	Ave.	0.594	3.0	2.531	0.013
20°C	1	0.594	3.0	19.647	0.098
	2	0.629	3.1	19.647	0.098
	3	0.612	3.1	17.050	0.085
	Ave.	0.612	3.1	18.781	00.94

Table 91 (*Continued*)

Temp.	No.	Weight of seed		Absorption of saccharose (mg)	
		200 seeds	1 seed Average (mg)	Per 200 seeds	Per 1 seed
30°C	1	0.614	3.1	16.916	0.085
	2	0.572	2.9	15.185	0.076
	3	0.568	2.8	15.052	0.075
	Ave.	0.585	2.9	15.718	0.079
40°C	1	0.618	3.1	12.055	0.060
	2	0.594	3.0	12.388	0.062
	3	0.552	3.0	10.589	0.053
	Ave.	0.588	3.0	11.677	0.058

The data in Table 91 show that the order of absorption of saccharose in relation to temperature was as follows: at 20°C>30°C>40°C>10°C. That is, the absorption of saccharose was largest at 20°C and least at 10°C. It is a noticeable fact that the absorption at 40°C was smaller than that at 30°C. Therefore, in this case, the permeability of seed coat and the exosmosis of seed materials may vary more conspicuously at the higher temperature than at the lower temperature. From these facts and the results shown in Tables 39, 41, 85 and 86 (Exp. 45), it may be considered that the retardation of the germination of *Picea* seeds at a higher temperature is due to the abnormal permeability of the external seed coat and the exosmosis of resorbed materials from the seeds.

2 Glycerine

Experiment 49

The concentration of glycerine in the present experiment was 0.1 mol.

Results as shown in Table 92 indicate that the absorption of glycerine by *P. Glehnii* seeds in the lower concentration was inferior to that in the higher concentration. The less the concentration of glycerine, the smaller was the absorption of glycerine. Such a tendency of absorption in the case of glycerine was exactly the same as that in saccharose solution. Next, experiment 50 was performed in order to study the influence of temperature on the absorption of glycerine.

Table 92

Absorption of glycerine by the seeds of *Picea Glehnii* in various concentration of glycerine.

Duration of experiment: Feb. 20–22. 1941.

Concentration of glycerine (mol)	Weight of seed.		Absorption of glycerine (mg)	
	200 seeds (grm)	1 seed (mg)	Per 200 seeds	Per 1 seed
1.0	0.588	0.0029	146.11	0.73
0.7	0.612	0.0030	69.33	0.34
0.5	0.608	0.0030	35.66	0.19
0.3	0.598	0.0029	52.00	0.24
0.1	0.622	0.0031	5.33	0.04

Experiment 50

The concentration of glycerine solution in the present experiment was 0.1 mol. Results are shown in Table 93.

The experimental results (Table 93) show that the absorption of glycerine at 20°C was always superior to that at any other temperature, and that it was less at 10°C than at 40°C and 30°C. The tendency in absorption of glycerine in this experiment resembled that of saccharose at various temperatures. In this case, also, the relation between absorption of glycerine and temperature may suggest a connection of the permeability of glycerine to the seed itself. In other words, the permeability or absorption capacity of seed varies with the changing of temperature. It is also shown that glycerine is more easily absorbable by seed than saccharose. From this fact it may be said that the temperature neighbouring 20°C is most suitable for the performance of the normal physiological function of *Picea* seeds.

Table 93

Absorption of glycerine by the seeds of *Picea Glehnii* at various temperatures.
Duration of experiment: Feb. 5–Feb. 8. 1941.

Temp.	No.	Weight of seed		Absorption of glycerine (mg)	
		200 seeds (grm)	1 seed Average (mg)	Per 200 seeds	Per 1 seed
10°C	1	0.6120	0.0030	26	0.13
	2	0.6270	0.0031	38	0.19
	3	0.6310	0.0031	32	0.16
	Aver.	0.6230	0.0031	32	0.16
20°C	1	0.5920	0.0029	38	0.19
	2	0.6120	0.0030	38	0.19
	3	0.6110	0.0030	34	0.17
	Aver.	0.6050	0.0030	36	0.18
30°C	1	0.5570	0.0027	60	0.30
	2	0.6020	0.0030	56	0.28
	3	0.5820	0.0029	56	0.28
	Aver.	0.5800	0.0028	57	0.29
40°C	1	0.5420	0.0027	50	0.25
	2	0.5870	0.0049	52	0.26
	3	0.5700	0.0028	48	0.24
	Aver.	0.5660	0.0034	50	0.25

3 Urea

As already mentioned, the influence of urea upon germination was very significant. Therefore, in order to find out the cause of the reduction in germination and relations between temperature and absorption of urea by seed the following experiments, Numbers 51 and 52, were carried out.

The results shown in these tables are the average of triplicate experiments.

Experiment 51

Results are shown in Tables 94 and 95.

Table 94

Absorption of urea by the seeds of *P. Glehnii* in various concentrations of urea.
Duration of experiment: Jan. 25–29. 1940.

Concentration of urea (mol)	Weight of seed		Absorption of urea	
	200 seeds (g)	1 seed (mg)	Per 200 seeds	Per 1 seed
0.01	0.582	2.9	0.6	0.003
0.05	0.574	2.9	4.0	0.020
0.10	0.610	3.0	5.2	0.026
0.30	0.602	3.0	16.0	0.799
0.50	0.599	3.0	82.5	0.413

Table 95

Absorption of urea by the seeds of *Picea jezoensis* in various concentrations of urea.
Duration of experiment: Jan. 25–29. 1940.

Concentration of urea (mol)	Weight of seed		Absorption of urea	
	200 seeds (g)	1 seed (mg)	Per 200 seeds	Per 1 seed
0.01	0.590	2.8	1.9	0.001
0.05	0.535	2.6	0.8	0.004
0.10	0.591	2.9	1.3	0.006
0.30	0.608	3.0	7.7	0.038
0.50	0.588	2.9	9.0	0.045

The data (Table 94) show that the amount of urea absorption from its solution of various concentration by the seeds varied with differences of the concentration. A significantly greater absorption of urea was detected from the solution of higher concentration than from lower concentration. Such a tendency was the same in the seeds of both *P. Glehnii* and *P. jezoensis*. The relation between the absorption of urea by seed and the concentration of its solution agrees completely with the present writer's results obtained by using root system of *Zea Mays* or storage tissue of *Brassica* sp. (YAMAGUCHI 1929). The absorption of urea per seed from urea solution of 0.3 or 0.5 mol concentration was significantly great in comparison with that of glycerine or saccharose. The greater absorption of urea may be responsible for the smaller percentage of germination of *P. Glehnii* and *P. jezoensis* seeds.

Experiment 52

This experiment was carried out to ascertain the influence of temperature on the absorption of urea by the seeds. Results are shown in Table 96.

Table 96

Absorption of urea by the seeds of *P. Glehnii* at various temperatures.
Duration of experiment: Feb. 10–Feb. 14. 1941.

Temp.	No.	Weight of seed		Absorption of urea (mg)	
		200 seeds (grm)	1 seed Average (mg)	Per 200 seeds	Per 1 seed
10°C	1	0.562	2.8	0.24	0.001
	2	0.584	2.9	0.30	0.002
	3	0.618	3.1	0.48	0.002
	Aver.	0.588	2.9	0.34	0.002
20°C	1	0.579	2.9	2.07	0.010
	2	0.609	3.0	2.07	0.010
	3	0.584	2.9	2.37	0.012
	Aver.	0.591	2.9	2.17	0.011

Table 96 (*Continued*)

Temp.	No.	Weight of seed		Absorption of urea (mg)	
		200 seeds (grm)	1 seed Average (mg)	Per 200 seeds	Per 1 seed
30°C	1	0.570	2.9	2.52	0.013
	2	0.562	2.8	2.97	0.015
	3	0.613	3.1	2.82	0.014
	Aver.	0.582	2.9	2.77	0.014
40°C	1	0.604	3.0	0.93	0.005
	2	0.599	3.0	1.08	0.005
	3	0.617	3.1	1.08	0.005
	Aver.	0.607	3.0	1.03	0.005

The order of urea absorption by *P. Glehnii* seed was as follows: 30°C>20°C>40°C>10°C. This order differs from that of saccharose absorption as shown in Table 89. But the absorption of urea or saccharose from corresponding solution was the least at 10°C. From the results shown in Tables 91, 93 and 96 it will be clearly seen that the absorption of urea, glycerine or saccharose by seed grains was smallest at low temperature, middling at a slightly higher temperature, and largest at moderate temperature such as about 20°C.

VI Suction force of seed during germination

The dormant seeds begin to absorb water and swell in the first step of germination, and then they germinate. In these processes the manner of absorption of water by seed-grain may be accepted to be analogous to the general cases of absorption of water by plant cells and tissues. Therefore, absorption of water by living cells may be touched upon briefly here in order fundamentally to understand the same by seeds.

The absorption of water by plant cells or tissues depends on their physico-chemical properties, namely, semi-permeability of protoplasmic membrane, composition of cell sap, and permeability and elasticity of cell wall. Cell sap is separated from the surrounding solution by two membranes, viz., cell membrane and protoplasmic membrane. They differ in their properties. Current theories regard that the protoplasmic membrane is semi-permeable, while the cell wall is permeable. As the cell wall possesses considerable toughness and elasticity, its resistance to internal pressure becomes greater when it is distended more, and when the counterbalance between them is reached the absorption of water and swelling of the cell ceases.

When the cell sap imbibes water, it increases in volume and expands the protoplast, which exerts pressure upon the cell wall. A counter-pressure on the cell content is produced by the cell, owing to its elasticity, acting in the direction opposite to that of the osmotic pressure. As the cell swells by absorbing water, the pressure of the cell wall on the cell contents increases.

If the osmotic pressure of the cell sap is denoted by P, and the pressure of the cell wall on its contents (wall pressure) by T, the condition of complete saturation is represented by $P=T$.

In this condition water neither enter nor escape from the cell, no matter how high the osmotic pressure inside the cell, this pressure being balanced by the counter-pressure of the distended cell wall. The cell in such condition is quite turgid.

In turgid condition, the "turgor pressure" of the cell is equal to the counter-pressure of the distended cell wall and protoplasmic membrane. In case the osmotic pressure of the cell sap is greater than the pressure of the elastic cell wall, if such an unsaturated cell is immersed in water or in dilute solution, this cell can absorb water, and a certain increase in the dimensions of the cell will occur. In such case, denote the water-absorbing force of the cell by S, then the following equation will be obtained;

$$P=T+S \quad \text{or} \quad S=P-T.$$

The value of S has been called "suction force" (Saugkraft) by URSPRUNG and BLUM (1916). But MAXIMOV (1925, p. 38–39) stated that "this term is not quite correct, as we are dealing not with a force, but with a pressure which can be expressed in atmospheres like osmotic pressure and turgor pressure, the difference between which it represents. We may therefore use the term "suction pressure" as suggested by STILES (1924)."

KOSTYTSCHEW and WENT (1931) said that the term "suction force" (Saugkraft) is not clear physically and is not correct, but as the term "suction force" has been commonly accepted in the field of plant physiology, they did not desire to adopt another term.

As mentioned above, though the term "suction force" is questioned by many authors, in the present paper the writer has used it. In the field of germination physiology the investigations on suction force of seed and especially in relation to the same problem in general plant cells or tissues have been rather rare. Concerning the "suction force" of seeds BUCHINGER, SCHRATZ and others have disagreed in their opinions, while in Japan several authors have followed BUCHINGER's interpretation, in spite of its being quite insufficient to the present author's belief. BUCHINGER (1927) determined the suction force of seed in accordance with the percentage of germination of seed. He regarded the mol concentration of the saccharose solution, in which the percentage of germination of seed is nearly a half of that in pure water as the suction force.

Some authors follow BUCHINGER exactly in determining the suction force of seed, but SCHRATZ (1932) said that BUCHINGER's measure is incorrect for obtaining the true or actual suction force of seed. In the present writer's opinion, this problem must be more strictly investigated on the ground of experimental results, because the BUCHINGER's method is too artificial to determine the water absorbing capacity of living seeds in reality.

The writer carried out the following experiments in order to study the suction force of dormant seeds or germinated seeds and to verify the relations between the suction force of plant cell and that of seed. Consideration was given to the present experimental results in comparison with suction force.

Experiment 53

The indirect method by plasmolysis, the equilibrium method and the direct weighing method were used for the determination of suction force of the cell. The weighing method, which depends on the change in weight of material, has been used for investigations in zoological physiology (HÖBER, 1926, p. 422). In plant physiological investigations STILES and JØRGENSEN (1919) employed the weighing method in determining the suction force of cut blocks of potato tubers.

In the present experiment the direct weighing method was adopted for determining the suction force of the seeds. The weight of germinated seeds, which were immersed in solutions of different concentrations are measured in order to see whether their weights have decreased or increased. At first, after the materials were weighed by torsion balance they were put into a glass vessel containing saccharose solution of definite concentration. These vessels were kept in the thermostat of 20°C for a certain number of hours, then the materials were taken out, and washed quickly with distilled water using a washing flask. The treated materials were laid on a sheet of filter paper and rolled once or twice to remove excess water and then were weighed again. All such processes were necessity quickly performed in order to avoid any experimental errors. The concentration of the solution, in which no change or the least change of weight was observed, was regarded as the value corresponding to the average suction force of the seed grains.

As experimental materials germinated seeds of *P. Glehnii* and *P. jezoensis* were used, and the suction force was determined of germinated seeds or seedlings at various germinating stages by the above described method. The results of this experiment are tabulated in the following tables.

The results in Tables 97–103 show the suction force of dormant seeds or various parts of germinated seeds. Table 103 was drafted by summarizing the data shown in Tables 97–102. From Table 103 it may be seen that the suction force of the dormant seed itself is enormously great, that is to say its value reaches at least 225 atm. p. presented by saturated sucrose solution at 20°C., and then the suction force of seed decreased with the advancing stage of germination. In other words the suction force of the dormant seeds commenced to drop with the beginning of germination. The suction force of slightly swollen seeds was 2.25 mol and that of slightly germinated seeds 1.30 or 1.15 mol; furthere, that of more germinated seeds decreased step by step with the advance of the germination stage and finally reached only 1.2 mol. Therefore, it is needful to distinguish precisely every stage of the germination process of seed when determining the suction force of dormant or germinated seeds. In general, the germination of a seed is considered to have occurred when the emerged radicle has reached a length of about 3 mm and began to curve downward. Then according to the writer's observation, the suction force of germinated seed of *Picea Glehnii* in the above described stage was found to be 1.25 mol of saccharose solution.

In the experiment, in which the plumula of germinated seedling was used and the results were shown in Table 100, it was found that the turgid plumulae softened obviously in saccharose solution of higher concentration than 1.1 mol, and the softening became more quick and severe in higher concentrations. In the solution of lower concentration such softening was not recognized. The softening of tissue in the present case may depend on the exosmosis of water from the cells owing to the higher concentration of the surrounding saccharose solution.

Table 97

Change of weight of dormant seeds, submerged 2 days in water, then kept 5 hours in saccharose solution at 20°C.

Concentration of saccharose (mol)		2.63	2.5	2.4	2.0	1.7
Increased or decreased weight of seed (mg)	*P. Glehnii*	+0.2	+0.2	+0.4	+0.2	+0.6
	P. jezoensis	+0.1	+0.2	+0.3	+0.5	+0.6

Table 98

Change of weight of slightly swelling seed, laid in thermostat of 20°C for 5 days supplied with moderate amount of water, then kept 5 hours in saccharose solution at 20°C.

Concentration of saccharose (mol)		2.63	2.5	2.4	1.7	1.5
Increased or decreased weight of seed (mg)	*P. Glehnii*	0	+0.2	+0.3	+0.2	+0.5
	P. jezoensis	0	+0.2	+0.3	+0.5	+0.5

Table 99

Change of slightly germinated seed, laid in thermostat of 20°C for 6 days supplied with moderate amount of water, then kept 5 hours in saccharose solution at 20°C.

Concentration of saccharose (mol)		1.5	1.4	1.3	1.2	1.1	1.0
Increased or decreased weight of seed (mg)	*P. Glehnii*	0	0	±0.1	±0.2	+0.2	+0.2
	P. jezoensis	0	0	0	±0.1	+0.2	+0.2

Table 100

Change of germinated seed, laid in thermostat of 20°C for 6 days supplied with moderate amount of water, then kept 5 hours in saccharose solution at 20°C. The length of the radicle was about 3 mm.

Concentration of saccharose (mol)		2.0	1.5	1.3	1.2	1.1	1.0	0.9
Increased or decreased weight of seed (mg)	*P. Glehnii*	0	0	+0.1	+0.1	+0.2	+0.2	+0.2
	P. jezoensis	0	0	0	+0.1	+0.1	+0.2	+0.2

Table 101

Change of weight of seedling, which germinated in the thermostat of 20°C, then was kept 5 hours in saccharose solution at 20°C. The length of the radicle was about 1.5–1.8 mm.

Concentration of saccharose (mol)		2.0	1.8	1.6	1.4	1.2	1.1	1.0	0.9	0.8	0.7	0.6	0.5	H_2O
Increased or decreased weight of seed (mg)	*P. Glehnii*	−1.1	−0.7	−0.3	−0.1	−0.1	−0.6	+0.6	+0.6	+0.4	+0.3	+0.3	+0.5	+0.8
	P. jezoensis	−1.1	−0.2	±0	−0.1	−0.7	−0.2	+0.3	+0.1	+0.1	+0.5	+0.2	+0.5	+0.5

Table 102

Change of stem in seedling, which germinated in the thermostat of 20°C, then was kept 5 hours in saccharose solution at 20°C. The length of the plumule was about 2.0–2.3 cm.

Concentration of saccharose (mol)		2.0	1.8	1.6	1.4	1.2	1.1	1.0	0.9	0.8	0.7	0.6	0.5	H_2O
Increased or decreased weight of seed (mg)	*P. Glehnii*	−2.8	−2.0	−1.0	−1.5	−1.4	±0	+0.3	+0.1	+0.2	+0.2	+0.2	+0.3	+0.3
	P. jezoensis	−1.1	−1.2	−1.6	−1.2	−0.8	±0	+0.8	+0.2	+0.2	+0.3	+0.2	+0.2	+0.4

Table 103

Summarized results of Experiment 50.

Germinating stage of seed	Suction force in Mol (cane sugar)	
	P. Glehnii	*P. jezoensis*
Dormant seed	2.7<	2.7<
Swelling seed	2.55	2.50
Cracked seed	1.80	1.60
Slightly germinated seed	1.30	1.15
Germinated seed	1.25	1.10
Radicle of germinated seed	1.05	1.05
Plumule of germinated seed	1.10	1.10

The phenomenon of water absorption by dormant or germinated seeds submerged in saccharose solutions of various concentrations is comparable with that of plant cells, because a swelling seed may be regarded as an osmometer. SHULL (1916) determined the water retaining capacity of soil by using *Xanthium* seeds regarding the seed as a living osmometer, because the seed of *Xanthium pennsylvanicum* possesses a semi-permeable coat, swells quickly and becomes isotonic with the surrounding solution.

Then, it may be said that the determination of suction force by direct weighing method applied by the writer is reasonable in order to ascertain the suction force of dormant or germinated seeds. But, as already mentioned, BUCHINGER (1927) proposed another method for this purpose. If the suction force of the seeds of *P. Glehnii* and *P. jezoensis* were calculated according to BUCHINGER's system based upon the writer's experimental results shown in Tables 59–62, it should be only 0.25 mol and 0.18 mol. These values were nearly in agreement with those given by TAZOE (1937), who reported that the suction forces of the seeds of *P. Glehnii* and *P. jezoensis* were 0.265 mol and 0.175 mol according to BUCHINGER's system.

The suction force of these *Picea* seeds obtained by BUCHINGER's system was much lower in comparison with that of the writer's results determined by the direct weighing method as shown in Table 103. The term "suction force" should be used in the same meaning either in the case of ordinary plant cells or tissues or in the case of seeds. The writer determined the suction force of *Picea* seeds by direct weighing method from the standpoint of plant physiology, but when it was calculated by BUCHINGER's method based upon the same data, the values were quite different. There is a question regarding not only the method but also regarding BUCHINGER's idea of "suction force". In this connection several authors have already expressed their disagreement in opinion with BUCHINGER. BERKNER and SCHLIMM (1929) and MEYER (1929) said that BUCHINGER ought to use "Keimenergiebestimmungen in Rohrzucker oder Keimprüfungen" instead of the term "Saugkraftbestimmungen". SCHRATZ (1932) also pointed out the unreasonableness of regarding the value of "suction force" of seed determined by BUCHINGER's system as a true suction force.

BUCHINGER (1933), however, made a protest stating that he never used the term "suction force" of seed in the same sense as "suction force" of plant cell. If this is so, it seems better to use some other term than "suction force" ("Saugkraft") in order to avoid misunderstanding.

VIII The change of sugar in germinated seed

During seed germination the mobilization occurs of storage substances such as carbohydrate, protein, oil and fat. Among others, the change of carbohydrate is one of the most important, because carbohydrate is used for energy metabolism of seedling. Investigations of these problems have been made by some authors[1] from the view point of agricultural science using agricultural seeds. But, in the field of forestry science few studies in this connection have been published up to the present time. The writer carried out the following experiment to learn about the change of sugar in *Picea* seeds during their germination process.

Experiment 54

Method of experiment: Reducing sugar and total sugar in a definite

1) CZAPEK (1920), KOSTYTSCHEW (1926), LEHMANN und AICHELE (1931), MILLER (1931), and MALHOTRA (1933).

number of seed grains in various stages of germination were estimated by the method described in chapter VI of this paper. Results are shown in Table 104.

Table, 104

Average amount of reducing sugar and total sugar in 100 grains of germinated seeds.
Duration of experiment: Jan. 30–Feb. 3. 1940.

Day kept in germination dish	*P. Glehnii*		*P. jezoensis*	
	Red. sugar	Total sugar	Red. sugar	Total sugar
4	0.53 mg	4.22 mg	0.47 mg	2.44 mg
6	0.60	6.66	0.60	6.57
7	0.73	6.11	0.93	4.22
8	0.47	2.45	0.40	2.44
9	0.33	2.11	0.07	1.88
10	0.26	2.22	0.07	1.67

The results shown in Table 104 indicate clearly that the content of reducing sugar and total sugar in germinated seed varied with the change of germination stage. The amount of reducing sugar being contained in 100 seeds of *P. Glehnii* or *P. jezoensis* was very much larger at the beginning of germination than in the later stage, and also the content of total sugar become smaller concomitantly with the progress of germination. Then such consumption of sugar may be the result of seed respiration during germination.

From Table 104 it will be seen that the total sugar content on the 6th day of germination was much larger than at any other stage. From such facts it may be considered that the total sugars including non-reducing sugar may be formed by decomposition of starch as a result of the action of amylase. In the present study the change of amylase action corresponding to the germination stages was investigated in the following manner. Action of amylase was estimated by the method of BACH and OPARIN (1922) as follows: the seeds were first divided into three groups, namely dormant, swollen and germinated groups. One hundred grains of each seed group were separately ground in porcelain mortars, and added with 20 cc of water. Thus prepared gruel was left to stand for 5 minutes, and then filtered. Filtrate was prepared from each seed group, and it was put into an ERLENMEYER's flask of ca 50 cc capacity with other substances.

Extract of sample	5 cc
Starch (1 %)	10 cc
Phosphate buffer mixture	10 cc
Thymol	a small piece

The pH-value of this mixed solution was 5.6 and remained unchanged during the experiment. After keeping this mixture for 24 hours in the thermostat of 40°C, the total reducing sugar with maltose was estimated by the method described in Chapter VI, and the amylase action was expressed by the amount of reducing sugar. Results are shown in Table 105.

Table 105

Amount of reducing sugar with maltose decomposed by amylase (mg. ±0.0005)
Duration of experiment: Feb. 2–4. 1940.

Material	*P. Glehnii* 100 grains	*P. jezoensis* 100 grains
Dormant seed		0
Swollen seed	0.008	0.002
Germinated seed	1.200	0.028

It will be seen in Table 105 that the occurrence of amylase in *Picea* seeds varies greatly according to the stage of germination; it is contained to a large amount especially in the germinated seeds, and the amount of amylase in *P. Glehnii* seeds was larger than in those of *P. jezoensis*. But the action of amylase was not found in the dormant seeds themselves which did not absorb water and did not yet swell. Therefore, from such fact it may be said that the increase of total sugar in 6th day of germination of both *Picea* seeds (Table 104) may depend absolutely upon the action of amylase. Then it may be considered that the catalytic action of amylase causes the decomposition of starch, and in turn, such decomposition of starch may result in the decrease of the total dry weight of germinated seed accompanied with the consumption of reducing sugar and total sugar. Then in order to ascertain the weight change of seed during germination the following experiment was carried out.

Experiment 55

Seeds were divided into four groups, and the raw and dry weights of 100 grains of each sort of *Picea* seeds were taken according to their respective sorts. Results are shown in Tables 106 and 107.

Table 106

Change in average weight 100 germinating seeds of *Picea Glehnii.*
Duration of experiment: Feb. 2–12. 1940.

Material	Raw weight (g)	Dry weight	Difference of dry weight	
			Absolute	%
Dormant seeds	0.4031	0.3609	0	0
Swollen seeds	0.4510	0.3513	0.0096	2.73
Cracked seeds	0.4925	0.3406	0.0203	5.77
Germinated seeds	0.5708	0.3116	0.0493	14.03

Table 107

Change in average weight of 100 germinating seeds of *Picea jezoensis.*
Duration of experiment: Feb. 2–12. 1940.

Material	Raw weight (g)	Dry weight	Difference of dry weight	
			Absolute	%
Dormant seeds	0.2185	0.2071	0	0
Swollen seeds	0.2306	0.2060	0.0071	3.44
Cracked seeds	0.3125	0.1895	0.0176	8.54
Germinated seeds	0.3265	0.1866	0.0205	9.95

The quantitative data shown in Tables 106 and 107 denote the noticeable change of raw or dry weight of the seeds. The marked decrease of dry weight and increase of raw weight in the germinated seeds were clearly recognized. The former may be due to the consumption of carbohydrates by seed respiration and the latter may be due to the absorption of water by germinated seed. Then it is reasonable to consider that the decrease of dry weight in germinated seeds may be closely related with the loss of total weight of them by germination.

IX The germination capacity of seed and factors relating to the preservation of germination power

Germination power of seed begins with maturity after the fertilization, and reaches its maximum with an elapse of days. It is quite lost during long storage, though the velocity of loss of germination power varies according to the kind of seed and to other factors. Moreover, the period of retention of germination power depends on the maturity, seed character, temperature, water, oxygen, drying and physical and chemical treatments. Concerning these factors the following experiments was performed with seeds of *P. jezoensis* in order to study the relations between the germination power and maturity.

Experiment 56

Picea jezoensis seeds only was used, which are collected in the Teshio Experimental Forest of Hokkaido Imperial University. In 1939 and 1940 the yield of seeds of *P. Glehnii* was extremely poor owing possibly to unfavourable climatic conditions of these years, and the seeds were not available. The germination test was made by the method described under Experiment 21. Results are shown in Table 108.

The percentage of germination shown in Table 108 was very small in comparison with that shown in Table 40, because the seeds used in the present experiment were collected in the preceding autumn (1939). In general, the character of the seed varies widely with the difference of year of production. From Table 108 it will be seen that the germination power of *P. jezoensis* seeds gathered on Sept. 5 was equal to that of the seeds gathered on Oct. 5 or Oct. 15, and that some seeds gathered on Aug. 5, were very poor and slow in germination. Considering the percentage of germination of the seeds in the present experiment it may be said that

Table 108

Germination record of the seeds of *Picea jezoensis* dried at 20°C for varioua temes.
Duration of experiment: Mar. 5–Mar. 22. 1940.

Day gathered	Number of germinated seed after											Percentage of germination
	6 days	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–19 days	Total	
July 25, 1939	0	0	0	0	0	0	0	0	0	0	0	0
Aug. 5, 1939	0	0	0	0	0	0	0	0	0	6	6	6
Aug. 15, 1939	0	0	0	0	4	4	5	4	1	8	26	26
Aug. 25, 1939	0	0	0	0	0	1	3	3	5	9	21	21
Sep. 5, 1939	0	0	0	0	0	4	10	12	5	9	40	40
Sep. 15, 1939	0	1	4	11	11	8	4	4	1	2	46	43
Sep. 25, 1939	0	1	8	11	12	11	1	0	0	1	45	45
Oct. 5, 1939	1	2	6	9	9	12	1	1	1	1	41	41
Oct. 15, 1939	0	0	5	9	7	6	3	4	2	5	42	42

the maturity of *Picea* seed was completed by Sept. 5, and that the seeds gathered after Sept. 5 possessed good germination power. That is to say, the *P. jezoensis* seed has reached its full maturity at about 90 days after fertilization or pollination, and furthermore, some seeds acquired germination power, though it was incomplete, as early as about 70 days after pollination. Therefore the *P. jezoensis* seeds should be gathered on Sept. 5 or afterwards from this time on, and then the germination power will be preserved for a long time if the treatment of seed, such as drying and temperature, were carefully taken into consideration. To study the relations between the drying temperature and germination capacity the following experiment was carried out.

Experiment 57

The seeds of *P. Glehnii* and *P. jezoensis* were dried in drying thermostat of definite temperature for certain hours, and the water content of seed and germination power were examined. Results are shown in Tables 109 and 110.

Table 109

Germination record of the seeds of *Picea Glehnii*.
Duration of experiment: Oct. 15–Nov. 4. 1938. Temp. at 20°C.

Temp. C	Time (hours)	Water lost raw weight (%)	Number of germinated seed after 5 days	6 "	7 "	8 "	9 "	10 "	11 "	12 "	13–22 "	Total	Percentage of germination
40°	5	4.8	4	18	19	13	11	13	6	5	3	92	92
	10	5.3	2	6	11	19	29	6	14	2	8	91	91
	20	6.1	0	6	13	12	7	20	9	10	13	90	90
	30	5.5	0	4	6	7	4	8	18	6	36	89	89
50°	5	5.9	1	13	9	7	10	17	5	11	13	86	86
	10	5.8	4	10	17	21	12	8	7	5	8	92	92
	20	6.0	1	1	3	2	4	14	11	7	42	85	85

Table 110

Germination record of the seeds of *Picea jezoensis*.
Duration of experiment: Oct. 15–Nov. 4, 1938.

Temp. C	Time (hours)	Water lost raw weight (%)	Number of germinated seed after 5 days	6 "	7 "	8 "	9 "	10 "	11 "	12 "	13–22 "	Total	Percentage of germination
40°	5	4.2	7	11	18	13	12	7	4	2	4	78	78
	10	6.2	7	0	31	8	6	2	3	1	1	59	59
	20	6.6	0	22	22	14	5	6	2	2	1	74	74
	30	5.6	0	16	28	10	6	3	3	0	4	70	70
50°	5	5.9	4	32	15	9	4	3	2	2	7	78	78
	10	6.2	13	14	18	11	6	4	3	1	3	73	73
	20	6.7	6	17	16	6	6	7	3	3	6	70	70

The data shown in Tables 109 and 110 indicate that the seed dried at comparatively high temperature for long periods of time lost more water than these dried at lower temperature for shorter hours, but drying at higher temperature for longer hours does not result in so remarkable dropping of germination power of seed to compare with those dried at lower temperature for fewer hours. It may be said that the drying of the seeds at 40°C or 50°C for 2–5 hours is adequate for drying of both sorts of *Picea* seeds for the purpose of long storage of them. In general, the moderate drying of seed is very beneficial for long preservation of germination power. SATO and the writer (1940) have proved that the germination power of *Populus Sieboldii* could be preserved for a long time by drying and storage at lower temperature.

Experiment 58

The purpose of this experiment was to study the change of germination power of both sorts of *Picea* seeds as influenced by the method of preservation. The seeds of *P. Glehnii* and *P. jezoensis* were stored in paper sack at ordinary room temperature (approximately −15°C+25C) and in the constantly low temperature chamber (approximately −15°C) for 2–3 years, and then germination tests were performed. Results are shown in Tables 111 and 112.

Table 111

Germination record of the seeds of *Picea Glehnii* at 20°C.

Datum of germi. test	Storage	Number of germinated seed after												Percentage of germination
		5 days	6 "	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–20 days	Total	
13. 10	Lab	4	18	18	13	11	13	6	5	1	1	1	91	91
14. 5	Low	0	5	6	8	9	14	13	12	7	7	4	85	85
14. 10	Low	0	3	6	7	8	13	14	12	7	6	5	81	81
15. 5	Low	0	0	4	6	11	15	12	8	5	4	0	65	65
15. 10	Low	0	2	3	6	10	13	11	8	5	2	0	60	60
16. 4. 24	Lab	0	0	1	5	5	7	10	5	5	3	9	50	50
16. 4. 24	Low	0	0	2	17	8	6	9	4	4	0	4	54	54

Table 112

Germination record of the seeds of *Picea jezoensis* at 20°C.

Datum of germi. test	Storage	Number of germinated seed after												Percentage of germination
		5 days	6 "	7 "	8 "	9 "	10 "	11 "	12 "	13 "	14 "	15–20 days	Total	
13. 10	Lab	7	11	18	13	11	7	2	1	1	1	2	74	74
14. 5	Low	0	1	4	9	16	14	14	5	4	3	2	72	72
14. 10	Low	0	1	3	8	16	15	13	5	4	2	0	70	70
15. 5	Low	0	0	2	5	19	13	14	4	4	2	0	63	63
15. 10	Low	0	0	1	6	17	13	12	6	4	0	0	59	59
16. 4. 24	Lab	0	0	0	1	6	2	11	4	2	3	4	33	33
16. 4. 24	Low	0	0	3	9	17	11	2	5	7	2	3	59	59

Tables 111 and 112 show that the germination power of *P. Glehnii* and *P. jezoensis* seeds dropped in accordance with the increase of days of storage, though the degree of dropping was not so great that the germination power was completely lost, and the seeds of *P. Glehnii* stored in an ordinary room or in constantly low temperature chamber for three years, still showed about 50 percent germination. But the degree of decrease in germination power in *P. Glehnii* seeds was not the same as in those of *P. jezoensis*, and the influence of the storage temperature on the germination of the seeds of *P. jezoensis* was greater than that on *P. Glehnii* seeds. That is to say, the percentage of germination of *P. jezoensis* seeds stored in an ordinary room was 33 %, those of stored in a constantly low temperature chamber was 59 %, while germination of *P. Glehnii* was about 50 % in both cases respectively. What is the cause of such a difference in the drop of germination power between *P. Glehnii* and *P. jezoensis*? As one of the cause of this difference in the drop of germination power the size of seed may be taken into consideration. This idea seems to be not so unreasonable, because, according to SATO and the writer's experiment (1940) seeds with *Populus Sieboldii*[1] stored in a constantly low temperature

1) The size of *Populus Sieboldii* seed is very small, and the average number of seed grains was about 4 million per 1 litre.

chamber continued their germination power for 300 days and the percentage of germination was nearly 80 %, but the seed stored in an ordinary room completely lost their germination power after only about 40 days. From such an observation it may be said that the decomposition or change of stored substances in such small seeds may occur very easily, and in turn it may result in the decrease of germination power. As another cause, the vitality or the resistance of seeds to outer factors may be taken into consideration, because the smaller sized seeds was sensible to chemical or physical influences or to injurious substances which may be formed in the course of germination by decomposition of stored substances in the seed. For example, it was observed by the writer that under the same conditions the *P. Glehnii* seeds could germinate, while the *P. jezoensis* could not. It may be said from the following observations that the decomposition or change of stored substance may occur during long storage and it may result in the diminishing of germination power. *Picea* seeds which had beed stored for 3 years in a constantly low temperature chamber did not germinate in LiCl solution of various concentrations, though the same seeds germinated very well in water, while the seeds stored for 1 or 2 years in similar condition germinated in the same solutions.

It was proved that catalase is contained widely in seeds and some intimate relations exist between the action of catalase and the germination power of seed, maturity, after-ripening, seed respiration or storage period of seeds. Then, it is reasonable to consider that some relations may exist between the germination power of *Picea* seeds and catalase action in them. Therefore, catalase activity in seed was determined in order to study the relations between the action of catalase and the difference of stored temperature. Catalase in the seeds was estimated as follows. Two grams in raw weight of the material were ground in a porcelain mortar, and added with about 70 cc of water. Thus prepared gruel was filtered, and the filtrate was diluted to 100 cc with water using measuring flask. This filtrate was put in ERLENMYER's flask of ca 50 cc capacity with other substances :

Extract of sample	10 cc
Hydrogen peroxyde (1%)	5
H_2SO_4 (S.G. 1.08)	3

After keeping this mixture for 30 minutes at 20°C in laboratory, the mixed solution was boiled for 5 minutes, and then the hydrogen peroxide was titrated with $KMnO_4$ (N/10). As control, the mixed solution was boiled immediately after being prepared. Obtained results are shown in Table 113.

Table 113

Amount of $KMnO_4$ (N/10, cc) used for titration of H_2O_2.
Date of experiment: Apr. 23. 1941.

Material	Stored in laboratory	Stored in low temperature chamber	Difference
P. Glehnii	310	310	0
P. jezoensisi	211	310	99

From this table it may be seen that the catalase action in *P. Glehnii* seeds stored in different ways did not show any difference, but in *P. jezoensis* seeds it was different. That is to say the catalase activity in *P. jezoensis* seeds stored at constantly low temperature chamber was greater than that in seeds stored in an ordinary chamber, and the percentage of germination of the former seeds was larger also than that of the latter. These two facts suggest the existence of some relations between the change of reserve substances in seeds and the storage temperature of the seeds, and also the connection among the change of reserve substance, the action of catalase and germination capacity of the seeds at least in so far as the seeds are very small and the substance reserved in the seed is very poor.

X General discussion

The results obtained in the present experiments show that the germination of seed is a complex process comprising swelling, absorption of water, salts and other substances and change or mobilization of reserved substances in the seed. It depends a great deal upon the character and kind of seed and upon outer factors, such as water, temperature, salts and non-electrolytes. The salts which acted to accelerate the swelling of seeds also accelerated the absorption of water; on the contrary the salts, which retarded the swelling of seeds, diminished the absorption of water by them. Such action presented by the salts may be attributable to the ionic action which occurs as a result of their dissociation in solution. Such influences of ions or salts upon the absorption of water by seeds and their swelling should be attributed to the ionic action on the protoplasm of plant cell. Consequently the actions of salts on seeds were quite in resemblance with those on the plant cells or tissues other than seed. Phenomena analogous to the present cases have been reported by LOO (1931) on the absorption of ammonia and nitrate by the root system of higher plants in relation to the effect of other salts, by PIRSCHLE and MEGDEHL (1931) on absorption of salts by plants, by LUNDEGÅRDH (1932) on the relation between the action or resorption of ions and plant growth, by BAPTISTE (1935) on the effect of some cations on the permeability of cells to water, and also by KREUT (1941) on the influence of salts upon the permeability to other substances. Therefore it may be said that the electrolytes which accelerate the swelling of the protoplasm or softening the plasma membrane tend to increase the permeability of the seed coat, on the contrary the electrolytes such as $MgCl_2$, $CaCl_2$ which check the swelling of protoplasm tend to decrease the swelling of seeds and the absorption of water by them. But the influence of salts on the germination of *Picea* seeds differs from that on the absorption of water by them. For example, the salts which retard the absorption of water did not always cause a drop in the percentage of germination nor did the salts which accelerate the absorption of water always promote the germination of the seeds. In the salt solution of lower concentration, the action of ions or salts on the germination of seeds or absorption of water by them may depend on the ionic action of salts, and not on the osmotic relationship, but in the salt solution of higher concentration the action of salt on the germination or absorption of water depends upon the osmotic value of solution as well as the ionic action. In the experiment on the effect of combination of salts on the swelling and germination of the seeds, it was recognized that so-called antagonism of ions to the germination and to the absorption of water exists between K and Ca, or Na and Ca does not exist between K and Na. That is to say the action of one kind of ion was compensated or retarded by another sort of ion. Such antagonistic action of ions recognized in the present study fully resembles the results of several previous studies on the action of ions upon the protoplasm, plant cell or absorption of other ions or non-electrolytes published by BRENNER (1920), KAHHO (1924), SCARTH (1925), HANSTEEN-CRANNER (1919), YAMAGUCHI (1929), LOO (1931), PIRSCHLE (1232), BURSTROEM (1934) and HÖFLER (1940).

The influence of saccharose, glycerine, and urea on the seed-germination and the absorption of water differs in many points from that of electrolytes. The remarkable difference of the influences between electrolyte and non-electrolyte on the germination and the absorption of water is possibly due to the following: firstly, osmotic phenomenon; secondly, permeability; thirdly, ionic action; fourthly, physiological action. In the lower concentration, the influence of an electrolyte on the seed germination and absorption of water is attributable to its ionic action and permeability, but that of a non-electrolyte may be attributable to the permeability and physiological action. The actions of several kinds of non-electrolytes differ each other according to the kind and also to the concentration within the same compound.

In saccharose solution which does not enter easily into plant cell, plant tissue or seed grain, the osmotic pressure of solution is the most responsible factor affecting the germination or absorption of water. Therefore the value of suction force of seedling and the osmotic value of surrounding saccharose solution have much significance in the germination or absorption of water. The maximum concentrations of saccharose solution for germination of the seeds of *P. Glehnii* and *P. jezoensis* were 0.42 mol and 0.38 mol respectively. On the other hand, the suction force of slightly germinated *P. Glehnii* seeds was 1.30 mol and that of *P. jezoensis* was 1.15 mol, and then the suction force of seedling became smaller in accordance with the progress of the germinating process.

From the concentration of saccharose solution at which the incipient germination of the seeds took place the following conclusion may be reached: that the failure of germination in the seeds of *P. Glehnii* and *P. jezoensis* in saccharose solutions of 0.42 mol and 0.38 mol respectively may be caused by osmotical detriment, though the suction force of slightly germinated *P. Glehnii* seed was about 1.1 mol, because judging from the suction force of the germinated seeds of *P. Glehnii* it is probable that the seeds can not easily absorb water nor readily germinate in the saccharose solution of 0.42 mol concentration. But the relationships between glycerine or urea and the germination of the seeds of *P. Glehnii* and *P. jezoensis* are another thing to compare with the case of saccharose. That is to say, the two sorts of *Picea* seeds did not germinate in the urea solution of lower concentration than in the case of saccharose, while they both germinated much more easily in glycerine solution of higher concentration than in the case of saccharose. Then the osmotic explanation can no more be applicable to the cases of urea and glycerine as it was in the case of saccharose. As the cause of the difference of influence among urea, glycerine and saccharose on the germination of the seeds of *P. Glehnii* and *P. jezoensis*, the physiological action and the high permeability of these three non-electrolytes should be taken into consideration as described above. Urea permeates very easily into the seed grain, and consequently the injurious effects on the germination of seed and physiological function of plant will be caused not only by an accumulation of urea itself but also an extreme decrease of hydrogen-ion concentration and an accumulation of ammonia in the seed or so-called ammonia-poisoning. Owing to these three causes urea is very injurious to the germination of both *Picea* seeds, even when the concentration of urea was lower than that of saccharose in solutions. Though glycerine permeates very easily into seed grain like urea, it does not exert an injurious effect on the germination of the seed even when the concentration was much higher than the case of urea. As the cause of this diverse difference of urea and glycerine in the influence upon germination of the seeds the difference of their physiological action may be taken into consideration. Already HARRINGTON and CROCKER (1923) reported that urea of 1 normal concentration retarded the germination of wheat seed, while KLEBS (1889) observed that glycerine permeates into algae cells without any injurious effect, and WILLSTÄTTER and WALDSCHMIDT-LEITZ (1932) used glycerine as a substitute solvent for water in enzyme studies and got good results.

From the results obtained in the experiments of germination and of suction force of the seeds of *P. Glehnii* and *P. jezoensis* in glycerine solution of various concentrations it was concluded that the failure in the germination of seeds of *P. Glehnii* and *P. jezoensis* in glycerine solution of concentrations higher than 1.05 mol for the former species and 0.90 mol for the latter was caused by the high concentration, in other words high osmotic value of glycerine solution, because the critical concentrations of glycerine solution for germination of the *Picea* seeds were about equal to the value of suction force of germinated seeds.

Temperature exerts great influence on the germination of both kinds of *Picea* seeds together with the amount of water, and temperature influences not only the germination of seed, but also the exosmosis of substances from seed grain and absorption of non-electrolytes by the seeds. Then the relationships between germination and temperature may be attributable to the complicated relationship among the germination, temperature, exosmosis and permeability of the seed grains. In these connections it is understood that both *Picea* seeds are not injured but rather favoured by submerging under cold water for a long period. TOLMAN and STOUT (1940) reported that beneficial effect was exerted upon the germination of sugar-beet seeds by submerging them under water, and they said that the retarding or injurious substances exosmosed from seed grain may be removed by this treatment. This opinion is directly applicable to the *Picea* seeds submerged under water in natural forest. But EYSTER (1940) reported that soaking seeds in water caused the loss of reserved substances from them, and the resulting germinative capacity of the seeds was approximately inversely proportional to the amount of the loss.

According to the writers's experiment the suction force of *Picea* seeds was extremely great and its value decreased in accordance with the progress of germinating stages.

For the long preservation of seeds without injuring their germination capacity, the low temperature is the most important factor and then the size, species and contents of seed grain also are important. In connection with such factors occurs also the stopping, diminishing and accelerating of the respiration of seed and they effect many cases. Perhaps the intensity of anaerobic respiration in seeds during preservation may control the germination capacity, accompanied with change in action of enzyme and change in reserved substances. In conclusion it may be said that the temperature is the most important in its influence upon the germination capacity of seed, especially lower temperature is essential for the longer preservation of seed. Furthermore it may be said that the temperature plays a great rôle on the germination physiology, such as permeability, absorption and exosmosis, enzymic action and change of reserved substances together with the other physiological problems of seed such as vernalization and alternative temperature for the germination of seed.

XI Summary

1. Physiological studies on the germination of seeds of *Picea Glehnii* and *Picea jezoensis* were carried out concerning the absorption of water and non-electrolytes, the swelling, the suction force, the change of reserve substance, and the germination capacity.

2. Absorption of water by seed grains was also affected by the kind and concentration of inorganic salts, especially by the kind and valency of cations and by the pH-value of the solution. Absorption of water by seed in the solutions of glycerine, saccharose or urea depends upon the concentration and the permeability of the non-electrolytes to the seed grains.

3. The swelling of the seed grains of *P. Glehnii* and *Picea jezoensis* was affected by concentration or kind of salts and also by the combination of them. The percentage of swelling of the seed grains was larger in the width than in the length.

4. The concentration, kind, valency and combination of several kinds of chlorides play great rôles in the germination of both *Picea* seeds, and

also the germination of these seeds is influenced by the concentration and kind of non-electrolytes.

5. The salts which cause the greater absorption of water by seed grains do not always promote their germination. That is to say, the influences of salts on the absorption of water by seeds are not necessarily coincident with those on the germination of seeds. The antagonistic action of salts on the seed germination and absorption of water by seed was recognized, but it was not clear on the swelling of seeds.

6. The concentration of non-electrolytes, in other word, the osmotic pressure of a solution has a great effect on the seed germination. The critical concentration of saccharose for germination of the seeds of *P. Glehnii* is 0.42 mol and for that of *P. jezoensis* 0.38 mol. But, the critical concentrations of urea or glycerine solution for germination of *Picea* seeds are quite different from that of saccharose. In glycerine solution seeds germinated at a higher concentration than in saccharose solution, while in urea solution the seed germination was checked even at far lower concentration than in the case of saccharose.

7. The injurious effect of urea or the beneficial effect of glycerine upon the seed germination depend possibly on the difference of physiological action of every substance. That is to say, the injurious action of urea occur due to the accumulation of the same in seeds owing to its high permeability, and on the other hand due to the ammonia-poisoning caused by an accumulation of ammonia produced in consequence of the hydrolysis of urea by the action of urease which is slightly detected in germinated seeds, and the accumulated ammonia causes an extreme decline of the H-ion concentration in the seed to alkaline side. The beneficial effects of glycerine are due to its high permeability, and a substitution of glycerine for water occurs to some extent in seeds during the process of their germination.

8. The action of electrolytes upon seed germination depends upon the ionic action of cations or anions and molecules of salts so far as the concentration of electrolytes is dilute, while in the case of the non-electrolytes the action of saccharose depends on the osmotic pressure of its solution and that of urea and glycerine depends on their specific permeability and physiological action.

9. The germination of the *Picea* seeds was influenced by the hydrogen-ion concentration in case that the concentration of the used phosphate mixture solution was higher than 0.1 mol, while in dilute concentrations such effect was not seen. Therefore it was concluded that the influence of hydrogen-ion concentration upon seed germination is greatly influenced by

the concentration of phosphates or other salts contained in the solution.

10. The *P. Glehnii* seed possesses higher resistance to the unfavourable effects of the high concentration of germinating media containing various salts, glycerine, urea or saccharose in solution than the *P. jezoensis* seed. That is to say, the *P. Glehnii* seed could germinate under more unfavourable conditions than the *P. jezoensis* seeds.

11. Temperature plays an important rôle in the germination of both *Picea* seeds in close connection with the amount of water. A temperature higher than 25°C is not favourable for the germination of the seeds possibly due to the acceleration of exosmosis of substances from the seed grains and the change of permeability of seed membrane caused by high temperature. But, a temperature lower than 10°C exerts no injurious effect upon the germination capacity of the seeds which were submerged under such cold water for a long period. These facts suggest that the climatic conditions in Hokkaido are suitable for the development of the *Picea* forest. It is rather a common even that the *Picea* seeds are buried under snow and submerged in cold water for the duration of about 5 months without any decline of their germination capacity.

12. The absorption of saccharose, glycerine and urea by the seed grains is influenced by temperature and concentration. The most conspicuous absorption of these three kinds of non-electrolytes was observed at 20°C in comparison with such other temperatures as 10°, 30° or 40°C.

13. Occurrence of a small amount of urease was detected in germinated seed, and the action of urease was not always favourable for the germination of seeds, though from the nutritional point of view urease is indispensable in utilizing urea as a nutrient. On the contrary, if the concentration of urea is high it causes ammonia-poisoning and extreme decrease of the hydrogen-ion concentration of seed owing to an accumulation of ammonia produced in the decomposition of urea by urease. These two facts stated above enhance the injurious effect due to urea combined with its own deleterious action.

14. The suction force of dormant seeds and germinated seeds was estimated by the direct weighing method using a torsion balance. The suction force of germinated seed is comparatively high to compare with that of the critical concentration of saccharose solutions for seed germination. The suction force of seedling varies according to its parts and then the value of suction force decreases gradually with the progress of germination process. From the results obtained in the writer's experiments it may be said that so-called suction force estimated by BUCHINGER does not show

the real suction force of seed.

15. Decreases in reducing and total sugars and total dry weight take place concomitant with the processes of germination, and it was proved that the action of amylase plainly increases in the germinated seeds.

16. In *P. jezoensis* seeds the germination power commences at 70 days after fertilization, and it reaches maturity by 90 days.

17. Storing the *P. jezoensis* seeds at low temperature was proved to be the most suitable method for the long preservation of germination capacity of the seeds, but it was another matter in the case of *P. Glehnii* seeds.

18. The influence of temperature on the stored seeds has an intimate relation with the size of each seed grain, in other words, with the amount of reserved substances in the seed. The beneficial effect of low temperature on preservation of germination power for a long period may depend principally on controlling anaerobic respiration of seeds. The anaerobic respiration may cause some unfavourable changes in reserve substances of seeds, or the formation of deleterious substances within seeds with the consequent result of the retardation of the germination of seeds.

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