THE ACTION OF CAFFEINE UPON THE GERMINATION AND GROWTH OF SEEDS

By FRED RANSOM, M.D., *En.*, *Beit Research Fellow.*

*From the Pharmacological Laboratory, Cambridge
(Second Communication)*

(Received January 17th, 1912)

In the following communication an endeavour is made to throw light upon the function which caffeine exercises in plant economy. At present the reason for the storage of caffeine in certain parts of some plants is quite as unexplained as is the appearance of its allies, xanthine and hypoxanthine, in animal tissues. On the other hand, the fact that it is found chiefly in the seeds and in young and growing shoots suggests that it may have something to do with germination or with growth.

In a previous paper I have shown that 1 per cent. solution of caffeine has a marked effect in delaying or preventing germination in a considerable variety of seeds. The following experiments unreservedly confirm those results. There was, however, the possibility that by treating seeds with very dilute solutions of caffeine a stimulating effect might be produced on germination or on growth or on both. The settlement of this point was one object of the present investigation.

The method adopted was the following:—On the bottom of glass needle boxes a thin layer of absorbent cotton-wool was placed, over this a piece of fine muslin was laid and the whole moistened with 50 c.c. of caffeine solution or tapwater. The seeds were then scattered thinly upon the muslin. The needle glasses were closed with loosely-fitting glass lids and stood either in the laboratory at about 17° C. or in a thermostat regulated to 22° C. Usually a series of glasses were charged with caffeine solutions of varying strength, with tapwater controls. At certain intervals the growths were compared by examination with the eye and, finally, the seedlings were carefully removed, either with or without the ungerminated seeds, superficially dried on blotting paper and weighed at once.

As regards germination it was found that caffeine solutions of 1·0, 0·1, 0·01 per cent. caused very distinct delay. With 0·001 per cent. that was not so obvious, and seeds sown in 0·0001 per cent. germinated as quickly as the tapwater controls (Table I).

Besides delay, the stronger caffeine solutions had a very marked

inhibitory effect, so that of the seeds sown in 1.0, 0.1, and 0.01 per cent. fewer germinated than in the weaker solutions or in tapwater (Table II).

Very dilute solutions of caffeine, 0.0001 per cent. and less, were without any obvious effect at all, either on germination or on growth. As will be seen from the table, 0.001 per cent. appears to have a slight effect in hindering growth (Table III).

The effect of caffeine in solutions of about 0.01 per cent. or more, was such that the seedlings, weighed after two or three days' growth, were distinctly lighter than the tapwater controls (Table IV). In most of the following tables the same effect can be observed.

It being thus evident that caffeine added to tapwater does not stimulate germination or growth; further experiments were made to ascertain whether a different effect would follow when a salt or other substance was added to the caffeine solution.

NITRATE OF POTASH—SACCHAROSE. (All solutions in tap water)				
Cress 0.5 g. in each glass and 50 c.c. fluid. Sown on 6/12/1911. Room temperature				
Medium	Condition on		Condition on	Weight on
	8/12/1911		9/12/1911	12/12/1911
(1) KNO_3 0.05 % ...	... 1 and 4 are the best		Relatively about the same	2.620
(2) KNO_3 0.05 % ...	... 2 and 3 about equal			2.385
Caffeine 0.01 % ...	... but smaller than the others			
(3) Caffeine 0.01 % ...	...			2.590
(4) Tap water ...	...			2.830
(5) Saccharose 2 % ...	... 5 and 6 are about equal and less than 4		6 is smaller than 5. Both are less than 4	2.620
(6) Saccharose 2 % ...	...			2.350
Caffeine 0.01 % ...	...			

The table shows very distinctly the influence of caffeine in retarding growth, but there was no stimulation.

DEXTROSE. (All solutions in tap water)			
Mustard 1.0 g. seed in each glass and 50 c.c. fluid. Sown on 12/12/1911. Room temperature			
Medium	Condition on 14/12/1911		Weight on 16/12/1911
(1) Dextrose 0.8 % ...	... 5 and 6 are the best grown and are about equal		3.100
(2) Dextrose 0.2 % ...	...		3.730
(3) Dextrose 0.8 % ...	... 1 and 3 are the smallest		3.040
Caffeine 0.001 % ...	...		
(4) Dextrose 0.2 % ...	... 2 and 4 are about equal		3.670
Caffeine 0.001 % ...	...		
(5) Tap water ...	...		4.420
(6) Caffeine 0.001 % ...	...		4.320

There is here no evidence whatever that the caffeine combined with dextrose stimulates, on the contrary what little difference there is indicates retardation. The best growth took place in tapwater, followed closely by the 0.001 per cent. caffeine solution.

ALANIN. (All solutions in tap water)

Mustard 1.0 g. seed in each glass and 50 c.c. fluid. Sown on 16/12/1911. Room temperature

Medium	Condition on 19/12/1911 in order of merit	Number of not germinated 20/12/1911	Weight, 20/12/1911 without not germinated
Alanin 0.5 %	... Best	39	2.750
	Tap water		
Alanin 0.1 %	0.0001 % caffeine	27	3.465
Alanin 0.5 %	0.001 „ caffeine	62	2.265
Caffeine 0.01 %			
Alanin 0.5 %	{ 0.1 „ alanin	30	3.050
Caffeine 0.001 %	{ 0.0001 „ caffeine		
Alanin 0.5 %	{ 0.1 „ alanin	46	2.610
Caffeine 0.0001 %	{ 0.001 „ caffeine		
Alanin 0.1 %	equal { 0.1 „ alanin	27	3.190
Caffeine 0.01 %	{ 0.1 „ alanin		
	{ 0.01 „ caffeine		
	{ 0.01 „ caffeine		
Alanin 0.1 %		34	3.550
Caffeine 0.001 %			
Alanin 0.1 %	equal { 0.5 „ alanin	25	3.380
Caffeine 0.0001 %	{ 0.0001 „ caffeine		
	{ 0.5 „ alanin		
	{ 0.001 „ caffeine		
Caffeine 0.01 %		22	3.850
Caffeine 0.001 %	equal { 0.5 „ alanin	24	3.850
Caffeine 0.0001 %	{ 0.01 „ caffeine	25	4.355
	{ 0.5 „ alanin		
Tap water	... Worst	19	4.420

The table shows that more seeds germinated and the seedlings developed best in tapwater. There were no signs of stimulation, on the contrary the combination alanin-caffeine was more deleterious than caffeine alone.

WITTE'S PEPTONE. (All solutions in tap water)

Mustard 1.0 g. in each glass and 50 c.c. fluid. Sown on 19/12/1911. 22° C.

Medium	Condition on 20/12/1911	Condition on 21/12/1911	Weight on 21/12/1911
(1) Peptone 0.5 %	... 5 and 6 have begun to sprout	6 is much the best	3.410
(2) Peptone 0.5 %	...	Then 1 and 2	3.280
(3) Peptone 0.5 %	... 1, 2, 3, 4 have	Then 5	2.830
Caffeine 0.02 %	... hardly any sprouts		
(4) Peptone 0.5 %	...	3 and 4 are the least grown	2.700
Caffeine 0.02 %	...		
(5) Tap water	...		2.900
Caffeine 0.02 %	...		
(6) Tap water	...		4.800

The seeds in tapwater developed best, and the combination of peptone with caffeine was more inhibitory than either peptone or caffeine alone.

WITTE'S PEPTONE. (All solutions in tap water)

Mustard 1.0 g. in each glass and 50 c.c. fluid. Sown on 20/12/1911. 22° C.

Medium	Condition on 21/12/1911	Condition on 22/12/1911	Weighed on 22/12/1911	Arranged in order of merit
(1) Peptone 0.1 %	... 11 and 12 have germinated best.	12 is the best	3.220	Tap water, best
(2) Peptone 0.5 %	...	Then 9, 10, 11	2.960	0.0001 % caffeine
(3) Peptone 0.5 % Caffeine 0.01 %	... Then 9 and 10 ...	Of the rest, 1, 2, 8, 7, 6 are better than 3, 4, 5	2.820	0.01 caffeine 0.001 % caffeine 0.1 % P. + 0.0001 % caffeine
(4) Peptone 0.5 % Caffeine 0.001 %	... The rest are fairly equal, 5 being the least grown		2.930	0.1 % P. + 0.01 % caffeine
(5) Peptone 0.5 % Caffeine 0.0001 %	... %...		2.690	{ 0.1 % P. 0.1 % P. + 0.001 % caffeine
(6) Peptone 0.1 % Caffeine 0.01 %			3.370	0.5 % P.
(7) Peptone 0.1 % Caffeine 0.001 %			3.220	0.5 P. + 0.001 % caffeine
(8) Peptone 0.1 % Caffeine 0.0001 %	... %...		3.410	0.5 % P. + 0.01 % caffeine 0.5 % P. + 0.0001 % caffeine
(9) Caffeine 0.01 %	...		3.650	
(10) Caffeine 0.001 %	...		3.600	
(11) Caffeine 0.0001 %	...		3.840	
(12) Tap water	...		3.890	

The best results were obtained with tapwater followed closely by the weakest caffeine solution. The combination of caffeine with peptone was more inhibitory than either caffeine or peptone alone.

CONCLUSIONS

The foregoing experiments afford ample evidence that caffeine added in the proportion of from 1 per cent. to 0.01 per cent. to water, in which seeds are then sown, exercises a powerful effect in retarding germination and growth. If as much as 1 per cent. caffeine is present there may even be complete inhibition of germination.

On the other hand there is nothing at present to show that caffeine, either alone or in combination with other substances can act as a stimulant to plant life. Nevertheless, the subject is not exhausted and the investigations are being continued.

TABLE I

Sown on 31/10/1911, at Room temperature

Medium	Cress, 0.5 g. in each glass Condition on 3/11/1911	Radish, 18 seeds in each glass Condition on 4/11/1911	Mustard, 0.5 g. in each glass Condition on 3/11/1911	
1 % caffeine in water	... None germinated	None germinated	None germinated	
0.1 % caffeine in water	... A very few small shoots	None germinated	None germinated	
0.01 % caffeine in water	... The greater part sprouted 2 mm.	A few very small shoots	A few just showing	
0.001 % caffeine in water	... The greater part sprouted 3 mm.	The greater part sprouted 2 mm.	The greater part sprouted	
0.0001 % caffeine in water	} Equal. Almost all sprouted 3-4 mm.	Equal. Mostly sprouted 3 mm.	Equal. Almost all sprouted. The best grown	
Tap water only				... }
Mustard, 0.5 g. in each glass. Sown on 10/11/1911, at room temperature		Lettuce, 0.3 g. in each glass. Sown on 25/11/1911, at room temperature		Radish, 18 seeds in each glass. Sown on 5/12/1911. 22° C.
Medium	Condition on 13/11/1911	Medium	Condition on 28/11/1911	Condition on 7/12/1911
0.1 % caffeine	A few very small shoots	0.1 % caffeine	None germinated	A few very small shoots
0.02 % caffeine	Mostly sprouted, but small	0.01 % caffeine	Few and small	More and stronger
0.004 % caffeine	Mostly sprouted, longer	0.001 % caffeine	More and larger	More still and stronger
0.0008 % caffeine	} Mostly sprouted. The best grown. Equal	0.0001 % caffeine	} About equal. The best grown	About equal. Distinctly the best grown
Tap water only		... }		

TABLE II

Mustard, 0.5 g. in each glass. Sown on 31/10/1911. Room Temperature		Radish, 18 seeds in each glass. Sown on 5/12/1911. Room temperature		Radish, 18 seeds in each glass. Sown on 5/12/1911. 22° C.	
Medium	Number not germinated	Condition of seeds on 12/12/1911		Condition of seeds on 12/12/1911	
		Germinated	Not germinated	Germinated	Not germinated
1 % caffeine	... 103 = all sown	5	13	8	10
0.1 % caffeine	... 35	7	11	10	8
0.01 % caffeine	... 26	6	12	10	8
0.001 % caffeine	... 15	12	6	13	5
0.0001 % caffeine	... 17	14	4	13	5
Tap water	... 16	13	5	14	4

TABLE III

Mustard seed, 1.0 g. in each glass. Sown on 17/12/1911. 22° C.

Medium				Weight on 19/12/1911
0.001 % caffeine	...	...	...	4.520 g.
0.0001 % caffeine	...	...	...	4.750
0.00001 % caffeine	...	...	...	4.610
0.000001 % caffeine	...	...	...	4.770
Tap water	...	...	...	4.830
Tap water	...	...	...	4.710

TABLE IV

Turnip seed, 0.5 g. in each glass. Sown on 15/11/1911. Room temperature

Cress, 0.5 g. in each glass. Sown on 15/11/1911. Room temperature

Medium				Weight on 22/11/1911	Weight on 22/11/1911
0.1 % caffeine	...	...	...	2.400 g.	2.530 g.
0.02 % caffeine	...	...	...	3.070	3.830
0.004 % caffeine	...	...	...	3.530	4.320
0.0008 % caffeine	...	...	...	3.760	5.370
0.00016 % caffeine	...	...	...	3.740	6.320
Tap water	...	...	...	3.730	6.430