Chemistry. — "*Amygdalin as nutriment for Fusarium.*" By Dr. H. I. WATERMAN. (Communicated by Prof. Dr. J. BÖESEKEN).

(Communicated in the meeting of May 26, 1917).

A solution of amygdalin in tapwater which at the same time contained inorganic salts as NH_4NO_3, KH_2PO_4, and $MgSO_4$ remained in the laboratory for some time at the ordinary temperature. After 18 days spontaneous infection was observed. On the liquid a white flocky mass of mycelium had appeared, under which a rose-coloured underground.

From this mycelium something was transferred to a plate of malt agar and cultivated at the ordinary temperature. After 24 hours some growth could already be observed, after 2×24 hours a flocky mycelium had been formed, whilst 24 hours later a very vigorous development was observed. A white flocky mass of mycelium was visible then, the plate had obtained on some spots a yellow and on other spots a red colour. The red colour was especially concentrated in those places of the nutrient plate which in transferring had been in contact with the platinum-wire.

The following day (after 4×24 hours) the whole glass-box was filled up with white mycelium.

The microscopy of the thus isolated species of mould and especially the presence of sickle-shaped spores divided into several cells pointed to *Fusarium*.

This species of *Fusarium* developed well on nutrient soils of the composition: tapwater whether or not coagulated with agar and containing 2% amygdalin, 0,15% NH_4NO_3. 0.15% KH_2PO_4, 0,10% magnesiumsulfate (crystallised). From the means of isolation this could be expected.

On amygdalin-agar *Fusarium* developed as white flocky mycelium, whilst this nutrient plate was for the greater part coloured yellow.

Especially on this nutrient soil the formation of sickle-shaped spores came to the front.

Some days later the yellow colour had for the greater part become red, whilst the mycelium had shrivelled. Besides, on malt agar this shrivelling after a prolonged cultivation was observed too.

The formed red colour of the amygdalin plate did not dissolve

in boiling water. With hydrochloric acid the colour became yellow, with sodium hydroxide, ammonia or soda violet and otherwise. So the colour acted as indicator.

Furthermore I observed that the infection with *Fusarium* occurred too with solutions of amygdalin, on which *Aspergillus niger* had developed first (temperature 34°).

Twelve days after inoculation with *Aspergillus niger* I delivered the solutions in question containing amygdalin from the mould layer; the clear solutions were kept at ordinary temperature in tumblers covered with watch-glasses. Not all amygdalin was used, a part had remained.

Miss Prof. Dr. JOH. WESTERDIJK was kind enough to determine the isolated *Fusarium*. The found species was *Fusarium discolor var. triseptatum* [1]).

This organism was found first by SHERBAKOFF on rotting potatoes.

It was possible that in my case the descent would be the same, because a few years consecutively I had experimented on potatoes in the same laboratory.

With the isolated species of mould I made almost the same researches as some time ago with *Aspergillus niger*. [2])

TABLE I. *(Fusarium).*

Glucose as exclusive organic food.			*Amygdalin* as exclusive organic food.	
Composition of the culture liquid 50 cm³ of tapwater, in which dissolved 0.15 % NH_4NO_3, 0.15 % KH_2PO_4, 0.1 % magnesiumsulfate (crystallised). Ordinary temperature.				
A. 2 % glucose: 1000 milligr.			B. 2 % amygdalin: 1000 milligr.	
Assimilated glucose (milligr.)	Obtained dry weight of mould (mgr.).	Number of days after inoculation.	Assimilated amygdalin (milligr.).	Obtained dry weight of mould (milligr.).
1000	322	12	600	274
		15	not determined.	347
1000	297	73		
1000	300	220	not determined.	235
		230	not determined.	299

[1]) C. D. SHERBAKOFF, Fusaria of potatoes. Memoir N°. 6. Cornell University Agricultural Experiment Station, May 1915. p. 239.

[2]) Amygdalin as nutriment for *Aspergillus niger*, These Proceedings Vol. XIX, p. 922 (1917).

TABLE II *a*. (*Fusarium*).

Retarding influence of benzaldehyde and hydrogen cyanide.

Culture liquid: 50 cm³. tapwater, in which dissolved 0.15 % NH_4NO_3, 0.15 % KH_2PO_4, 0.1 % $MgSO_4$. 7 H_2O and 2 % glucose. Ordinary temperature.

No.	Added.	Development after				Obtained dry weight of mould (Mgr.) after 73 days.
		4	7	21	37 days.	
1	—	++	+++	+++++	+++++++	297 [2]
2	1 drop of benzaldehyde.	—	?	+ [1]	+++++	324 [2]
3	3 drops of benzaldehyde.	—	—	—	— [1]	
4	5 drops of benzaldehyde.	—	—	—	— [1]	
5	1 cM³ P	++	+++	+++++	+++++++	277 [2]
6	3 cM³ P	+	++	+++++	+++++	282 [2]
7	5 cM³ P	+	++	+++++	+++++++	238 [2]
8	5 cM³ Q	++	+++	+++++	+++++++	

The solution P was prepared as follows: 50 milligr. KCN was dissolved in distilled water and filled up to 100 cm³. Added 10 cm³ of 0,98 × $\frac{1}{10}$ Normal sulfuric acid. The solution Q was obtained by adding to 100 cm³ of H_2O 10 cm³ of 0,98 × $\frac{1}{10}$ Normal sulfuric acid.

TABLE II *b*. (*Fusarium*)

50 cm³. of tapwater, in which dissolved 0.15 % NH_4NO_3, 0.15 % KH_2PO_4, 0.1 % $MgSO_4$. 7 H_2O. Ordinary temperature.

No.	Added.	Development after		
		4	6	11 days
1,2	2 % glucose	++++	+++++++	+++++++
3,4	2 % glucose + 0,04 % emulsin	++	+++++	+++++++
5,6	2 % amygdalin	++, +++	+++++	+++++++
7,8	2% amygdalin+0.04% emulsin	— [3]	— [3]	— [3]

The results obtained with *Fusarium* were almost quite analogous with *Aspergillus niger*.

[1] Sweat smell of benzaldehyde could be stated.

[2] All glucose was used.

[3] The smell of benzaldehyde or (and) HCN was stated.

So it was proved:

1 Amygdalin is assimilated by *Fusarium* whilst young mycelium is formed at the expense of the assimilated amygdalin. (Table I).

2. Compared with glucose amygdalin is not an inferior nutriment at least with regard to the dry weight of mould obtained. (Table I).

3. Benzaldehyde and to a small degree HCN hinder the development of *Fusarium* in liquids containing glucose (Table II^a), whilst the addition of emulsin to liquids containing amygdalin prevents growth entirely. The same emulsin has practically no retarding influence on the development of *Fusarium* in glucose containing solutions (Table II^b).

Therefore it is impossible that when amygdalin as only source of carbon is assimilated by *Fusarium* this glucoside is dissociated to an important degree into glucose, benzaldehyde and HCN out of the cell.

Dordrecht, May 1917.