teries were not entirely occluded, the fall in pressure was much more rapid than in those instances (Group III), in which the arteries were absolutely occluded regardless of the condition of the veins.

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Liver Autolysis in the Peritoneal Cavity of the Dog.

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Mann observed in his experiments with hepatectomy in dogs that when small pieces of liver tissue were left behind in the peritoneal cavity, the dogs invariably died of a severe peritonitis in less than 24 hours. Mason[1] and coworkers confirmed Mann's observations, studied the blood chemistry in such dogs, and found that saline extracts of autolized dog's liver injected intravenously proved very toxic. If the liver was *not* autolized, no toxic reactions were observed. They described the typical picture of dogs dying following deposition of pieces of dog's liver into the peritoneal cavity. One to 300 cc. of a serosanguinous fluid was usually present and the peritoneal surfaces were reddened. After 24 hours, liver placed into the cavity could hardly be identified except as a friable mushy mass which contained gas. Wangensteen[2] showed the relative non-toxicity of rat's liver and adult dog's kidney compared to adult dog's liver. He also showed the existence of a quantitative tolerance of dog's liver introduced into the rat's peritoneal cavity, the rats usually not dying till 15 gm. of dog's liver per kilo body weight of rat had been exceeded. Ellis and Dragstedt[3] indicated that an anaerobic gas forming bacillus commonly found in the dog's liver was responsible for death in liver autolysis *in vivo*. They identified this organism closely with the Welch bacillus. Later Rewbridge[5] showed that introduction of sterile bile salts into the peritoneal cavity of dogs caused a peritonitis death similar to that of liver autolysis. Still

[1] Mason, E. C., Davidson, C. C., *et al.*, *J. Lab. and Clin. Med.*, 1924, **10**, 622, 906, 977.

[2] Wangensteen, O. H., *Endokrinol.*, 1928, **2**, 170.

[3] Ellis, J. C., and Dragstedt, L. R., *Arch. Surg.*, 1930, **20**, 8.

[4] Andrews, A., and Hrdina, L. S., PROC. SOC. EXP. BIOL. AND MED., 1930, **27**, 987.

[5] Rewbridge, A. G., *Surg. Gynec. and Obstet.*, 1931, **52**, 205.

later, Andrews[6] contended that the cause of death in liver autolysis was primarily due to a toxic agent extractable from fresh normal adult dog's liver and independent of bacterial activity. However, he claimed such deaths were accompanied by presence of Welch-like organisms in the peritoneal cavity at death.

The purpose of this paper is to repeat some of the work already done on the subject and attempt to determine more conclusively by the employment of filtrates of fresh liver tissue and liver tissue treated in various ways, as well as filtrates of cultures of the organisms found in liver tissue, whether the bacteria and their toxin are solely responsible for the death of the animal or whether other toxic products peculiar to liver tissue itself as contended by Andrews also plays a significant rôle in the death of the animal.

Method. Experimental samples of dog's liver, filtrates and ex-

TABLE I.

	Substance Introduced.	No. Dogs	Died	Survived
1	Rat liver	2	0	2
2	Adult dog kidney	1	0	1
3	" " liver—ground	4	4	0
4	Foetal dog liver—ground	1	0	1
5	Adult dog liver—ground—autoclaved (see Exp. 18 for similar result)	1	0	1
6	Adult dog liver—ground—boiled ½ minute	2	0	2
	" " 3 "	2	0	2
	" " 3 " —incubated	1	1	0
7	Broth culture dog liver anaerobes— 4 cc., 6 cc., 30 cc.	3	0	3
	—40 cc., 60 cc., 100 cc.	3	3	0
8	" " " " " —filtrate	1	1	0
9	" " " " " " —boiled	1	0	1
10	" media control—filtrate	3	0	3
11	" culture dog liver anaerobe—autoclaved liver—ground			
	2.5 cc. of culture— " " "	1	1	0
	15.0 cc. " " — " " "	1	1	0
12	Adult dog liver—ground—Aqueous extract—extract unfiltered	1	0	1
13	Adult dog liver—ground—Aqueous extract—extract filtered	4	0	4
14	Adult dog liver—ground—incubated—Aqueous extract—extract filtered—boiled	1	0	1
15	Adult dog liver—ground—incubated—Aqueous extract—extract unfiltered	1	1	0
16	Adult dog liver—ground—incubated—Aqueous extract—extract filtered	3	2	1
17	Foetal dog liver—ground—incubated—Aqueous extract—extract filtered	2	0	2
18	Adult dog liver—ground—autoclaved—plus filtrate fresh adult dog liver, combination incubated	3	0	3
19	Adult dog liver—ground—ether extract	2	0	2
20	" " " " —alcohol extract	4	0	4
21	Peritoneal fluid of dogs dying from liver autolysis—filtrate	2	0	2

[6] Andrews, A., Rewbridge, W. G., and Hrdina, L., *Surg. Gynec. and Obstet.*, 1931, **53**, 176.

tracts of such liver, and cultures of anerobic organisms were introduced into the peritoneal cavities of adult dogs. The samples of liver treated in the various ways described in Table I were introduced in amounts always equal to 4 gm. per kilo dog body weight. The filtrates and extracts were introduced in amounts equivalent to corresponding amounts of liver, namely 4 gm. per kilo. The amounts of bacterial culture introduced are given in the table. All filtrates made were Berkefeld filtrates. All extracts were either with physiologic saline, ether, or alcohol as specified. All bacterial cultures were approximately 18 hours old and were composed of gas forming anaerobic gram positive bacilli isolated from adult dog's liver. These organisms were used in pure form and were beyond question the same as those used by other workers in this field. All dogs reported having died in these experiments, died inside of 36 hours. Those that did not die lived several weeks when used for other purposes.

Results. Experiments 1, 2, and 4. As noted in the table, ground rat liver, foetal dog liver and adult dog kidney did not kill. These tissues were all cultured and found bacteriologically sterile.

Experiment 3. Ground adult dog liver invariably killed in the amounts specified. It always contained gas forming anaerobes.

Experiment 5. Autoclaved ground adult dog liver never killed. It was always sterile.

Experiment 6. Boiling ground fresh adult dog's liver probably kills vegetative forms but not spores of the anaerobes and therefore did not kill. Incubating the boiled liver perhaps liberates vegetative forms and therefore kills.

Experiment 7. At least 40 cc. suspension of the anaerobes alone was found necessary here to kill.

Experiments 8 and 9. Filtrate of such a suspension kills but not after being boiled, indicating presence of a thermolabile toxin.

Experiment 10. Filtrates of liver peptone broth similar to that in which the organisms were grown were introduced intraperitoneally as controls. They did not kill.

Experiment 11. Sublethal amounts of bacterial suspension plus autoclaved ground liver kills.

Experiments 12 and 13. Aqueous extracts of fresh ground adult dog's liver did not kill. It contained no bacterial toxin.

Experiments 14, 15, and 16. When the liver was first incubated, such extracts did kill, but again, not after boiling. This would point towards presence of a toxin produced by bacteria and destroyed by heat.

Experiment 17. Filtrates of incubated foetal liver did not kill. No bacteria were present in this liver in the beginning.

Experiment 18. When autoclaved ground adult dog liver plus filtrates of fresh ground adult dog liver was allowed to incubate the animals survived. This would show that water soluble enzymes from the fresh ground liver acting on autoclaved liver unaided by bacteria produced no toxic substance. This would point against the presence of a toxin produced by aseptic enzyme autolysis of liver.

Experiments 19 and 20. Ether and alcohol extracts of ground fresh adult dog's liver did not kill.

Experiment 21. Filtrates of peritoneal fluid from dogs dying of liver autolysis proved non-toxic as already shown by others. The toxins are already probably absorbed by the host.

Bile salts are not found in the amounts of dog liver used in these experiments in sufficient quantity to cause death.

Conclusions. 1. Anaerobic bacteria or their toxic products cause death in autolysis of dog's liver *in vivo*. 2. No toxic product was found in the liver responsible for death which was inherent to liver tissue and independent of bacterial activity.