Doche I.

X-13-16

Complement fixation c̄ Serum of rabbit injected c̄ pneumococcus No II.

1 cc Pn II. 44' 18 hr. both injected i.p. rabbit bled from ear before injection, 4 hrs after and 6½ hrs after. Blood allowed to clot in ice box Serum separated and passed thru a normal Berkfeld filter. Pneumococci in smears from blood 4 and 6½ hrs. after infection. Organisms comparatively scarce. Sheep cells used for haemolysis, antisheep rabbit amboceptor and G.P. complement.

Titration of amboceptor. Dil 1-500.

Ambo.	10% Comp.	Cells.	Result.
.5 cc	0.5 cc	0.5 cc	C.H
.4 cc	0.5 cc	0.5 cc	C.H
.3 cc	0.5 cc	0.5 cc	C.H
.2 cc	0.5 cc	0.5 cc	C.H
.1 cc	0.5 cc	0.5 cc	+
.5 cc	—	0.5 cc	0
—	1.0	0.5 cc	0
—	—	0.5 cc	0

X-13-16.

Anticomplementary action of Ser II Snowflake bot T.

Ser II	10% Comp.	1-500 Amb.	Cells.	Result.
0.5 cm	0.5 cm	0.5 cm	0.5 cm	+++
0.2 cm	0.5 cm	0.5 cm	0.5 cm	+
0.1 cm	0.5 cm	0.5 cm	0.5 cm	0
0.75 cm	0.5 cm	0.5 cm	0.5 cm	0
0.05 cm	0.5 cm	0.5 cm	0.5 cm	0
0.025 cm	0.5 cm	0.5 cm	0.5 cm	0
0.01 cm	0.5 cm	0.5 cm	0.5 cm	0
0.005 cm	0.5 cm	0.5 cm	0.5 cm	0
0.001 cm	0.5 cm	0.5 cm	0.5 cm	0
0.5	—	—	0.5 cm	++++
0.2	—	—	0.5 cm	++++
—	0.5 cm	—	0.5 cm	++++
—	—	1.0 cm	0.5 cm	++++
—	—	—	0.5 cm	++++

Serum inactivated at 56° C for 1/2 hr.

X-13-16.

Anticomplementary action of infected rabbit serum used as antigen in experiment.

Rab. Ser.	10% Comp.	Amb. 1-500	Cells.	Rb. Ser. 4 hrs.	Rb. Ser. 6 1/2 hrs.
0.5 cm	0.5 cm	0.4 cm	0.5 cm	++++	0
0.3 cm	0.5 cm	0.4 cm	0.5 cm	++++	0
0.1 cm	0.5 cm	0.4 cm	0.5 cm	++++	0
0.05 cm	0.5 cm	0.4 cm	0.5 cm	++++	0
0.5 cm	—	—	0.5 cm	0	++++
0.2 cm	—	—	0.5 cm	0	++++
—	0.5 cm	—	0.5 cm	0	++++
—	—	0.8 cm	0.5 cm	0	++++
—	—	—		0	

Rabbit Ser. not inactivated.

X-13-16. Complement Fixation with Serum of rabbit infected C̄ Pn II as antigen. Im. Ser. Pn II Serum plate Sol T.

Rb. Ser.	H. Ser II	10% Comp.	Amb 1-500	Cells.	Rb. S. 0 hrs.	Rb. S. 4 hrs.	Rb. S. 6½ hrs.
0.25"	0.05"	0.5"	0.4"	0.5"	0	++±	+++
0.2"	0.05"	0.5"	0.4"	0.5"	0	++±	+++
0.15"	0.05"	0.5"	0.4"	0.5"	0	++±	++±
0.1"	0.05"	0.5"	0.4"	0.5"	0	++±	++±
0.05"	0.05"	0.5"	0.4"	0.5"	0	++	++±
0.25"	—	0.5"	0.4"	0.5"	0	0	0
0.1"	—	0.5"	0.4"	0.5"	0	0	+
—	0.1"	—	—	0.5"	++++	++++	++++
—	—	0.5"	—	0.5"	++++	++++	++++
—	—	—	0.8"	0.5"	++++	++++	++++
—	—	—	—	0.5"	++++	++++	++++

Immune Ser. and Rabbit Ser. used as antigen inactivated at 56°C for ½ hr.

X-13-16.

Summary: Amboceptor and complement both very satisfactory. Haem olysis rapid and clear. Immune Horse serum not very anticomplementary 0.2" giving only + in bilirubin. Rabbit sera not anti-complementary nor haem olytic. 1/4 of the anticomplementary dose of Im. Ser. gives almost complete fixation in the higher doses of infected rabbit serum and partial fixation as low as 0.05" Rb. Ser. Rb. Ser. before infection shows no fixation nor anticomplementary action. Rabbit died in 24 hrs.

✓

XI-13-16. Occurrence of Soluble Material of Pneum. coccus in Human blood during Lobar Pneumonia.

Pt. O'Connor Wd iii. Organism Pn iii. Day of disease: Seventh. Blood culture +, 360 colonies per cc.

Precipitin reaction à blood serum.

0.5" Pt Ser. + 0.5" S i	–
0.5" Pt Ser. + 0.5" S ii	–
0.5" Pt Ser. + 0.5" S iii	+++

Precipitin reaction à urine.

0.5" urine + 0.5" S i	–
0.5" urine + 0.5" S ii	–
0.5" urine + 0.5" S iii	+++

Cultures from urine: Negative for Pn iii.

XI-13-16.

Complement fixation using Serum Pt. O'Connor as antigen and Antipn. Ser iii (Board of Health) as immune serum.

Serum O'Connor passed through a Berkfeld filter N. to free from organisms.

Antigenic and Im. Sera. inactivated at 56 °C. for 3/4 hr.

Antigen S. O'Connor	Im. Ser. Pn iii	Amb. Cepton	Comp. 10%	Cells 5% sus.	Result
0.2"	0.1"	2 units	0.4"	0.5"	++
0.1"	0.1"	"	0.4"	0.5"	+++±
0.05"	0.1"	"	0.4"	0.5"	+++±
0.01"	0.1"	"	0.4"	0.5"	++
0.4"	–	"	0.4"	0.5"	C.H.
0.2"	–	"	0.4"	0.5"	C.H.
0.2"	–	–	–	0.5"	0
–	0.2"	2 units	0.4"	0.5"	C.H.
–	0.2"	–	–	0.5"	0
0.2"	–	–	0.4"	0.5"	Sl.H.

Tubes made up to 2.5" c NaCl soc.

XI-13-16.

Titration of amboceptor and complement used in four grm experiment.

Amb.	Comp.	Cells.	Result.	Tubes in thermostat 1/2 hr. Sheep cells.
1-40.	1:10	5%		
0.5 cc	0.5 cc	0.5 cc	C.H.	Rabbit amboceptor.
0.4 cc	0.5 cc	0.5 cc	C.H.	S.P. complement.
0.3 cc	0.5 cc	0.5 cc	C.H	Amb unit 0.15 cc dil 1-40.
0.2 cc	0.5 cc	0.5 cc	C.H.	Comp. unit 0.2 cc 1:10.
0.15 cc	0.5 cc	0.5 cc	C.H.	
0.1 cc	0.5 cc	0.5 cc	Sl. im.	
0.5 cc	—	0.5 cc	0	
—	0.5 cc	0.5 cc	0	
—	—	0.5 cc	0	
0.3 cc	0.4 cc	0.5 cc	C.H.	
0.3 cc	0.3 cc	0.5 cc	C.H.	
0.3 cc	0.2 cc	0.5 cc	C.H.	

XI-13-16 ✓

Complement fixation using human Pt. A'Conn as antigen and Conf. pa. III (Bol. Gltenlth.) as immune serum.

Pt's serum Berkefeld. Sera in autoclave at 56° C for 45 min.

Antigen	Im. ser.	amboceptor	Comp.	Cells.	Result.
S. A'Conn.	Pa III		1:10	5% Sheep.	
0.01 cc	0.1 cc	2 units	0.4 cc	0.5 cc	+++±
0.008 cc	0.1 cc	"	0.4 cc	"	+++
0.006 cc	0.1 cc	"	0.4 cc	"	++
0.004 cc	0.1 cc	"	0.4 cc	"	+
0.002 cc	0.1 cc	"	0.4 cc	"	±
0.001 cc	0.1 cc	"	0.4 cc	"	−
0.01 cc	—	"	0.4 cc	"	−
—	0.2 cc	"	0.4 cc	"	−
0.01 cc	—	"	—	"	0
—	0.2 cc	—	—	"	0
—	—	4 units	—	"	0
—	—	—	—	"	0
—	—	—	1.0 cc	"	Sl. H.

XI-16-16.

Complement fixation using pneumonia patients

Serum as antigen. Pt Michael, pneumonia Type IV

Blood culture. Pt's and immune sera inactivated at 56°C.

Antigen Ser. Michael	Im. Sera Types.	Ambo-ceptor	Comp. 10%	Cells 5% Susp.	Result.
0.1"	0.1" N.H.S.	2 units	0.4"	0.5"	–
0.1"	0.1" S I	"	0.4"	0.5"	–
0.1"	0.1" S II	"	0.4"	0.5"	–
0.1"	0.1" S III	"	0.4"	0.5"	–
0.2"	—	"	0.4"	0.5"	–
0.2"	—		—	0.5"	0
–	0.2" N.H.S.	"	0.4"	0.5"	–
–	0.2" S I	"	0.4"	0.5"	–
–	0.2" S II	"	0.4"	0.5"	–
–	0.2" S III	"	0.4"	0.5"	–
–	—	4 units	—	0.5"	0
–	—	—	1.0"	0.5"	0
–	—	—	—	0.5"	0

Unit amboceptor = 0.15" dil 1-400. Unit Comp. = 0.2" 10% G.P. Ser.

XI-25-16. Comp. Fixation Serum Zimschitz Type III (hor. 22 + 25)

Pts Ser.	Im Ser.	Ambo.	Comp.	Cells.	Ser XI-22	Ser XI-25.
0.2	0.1 N.S.	2 units	0.5"	0.5"	–	–
0.2	0.1 S I	"	"	"	–	–
0.2	0.1 S II	"	"	"	–	–
0.2	0.15 S III	"	"	"	–	+
0.4	–	"	"	"	–	–
0.2	–	"	"	"	–	–
0.4	–	–	–	"	0	0
0.2	–	–	0.5"	"	C.H.	C.H.
–	0.2 N.S.	"	"	"	–	–
–	0.2 S I	"	"	"	–	–
–	0.2 S II	"	"	"	–	–
–	0.3 S III	"	"	"	–	–
–	–	4 units	–	"	0	
–	–	–	1.0"	"	Mod. H.	
–	–	–	–	"	0	

Comp. units 0.4" of 10% G. S. Serum.

Ambo unit 0.15" dil 1-400.

XI-17-16. Formation of Soluble material by Pn T during growth.

Culture D4.6". Planted in 50" plain broth + 10" ascitic fluid 1.0" of a 16 h. culture of D4.6³.

Culture plated at varying intervals and colonies counted.

Material centrifugalized and supernatant fluid used to titrate precipitin reaction.

Precipitin reaction titrated by diluting antigen (supernatant broth.) and adding 0.1" serum T to each tube. Total quantity in each tube 1.0".

Reaction read at end of 24 and 48 hrs. in ice box.

Supernatant fluids all heated at 56° C. for ½ hr.