Von Ostertag has steadily advocated preserving measly meat in a cooler for 21 days, but apart from those writers who pointed out the risk to public health, some of his other strong arguments have been negatived by several recent workers:

- (a) Clarenburg, who maintained a temperature just above freezing, found that putrefaction set in comparatively early. One feels that under commercial conditions in abattoirs, very rarely will it be practicable to preserve meat for such a period before putrefaction will do damage, in spite of von Ostertag's directions regarding the maintenance of low temperatures and humidity of the air, as we have already tabulated.
- (b) Von Ostertag's claim that under chilling conditions the least loss of weight will occur, has been refuted, practically, by Wagemann (1935), who staunchly preferred the freezing of measly carcasses to the uncertain 21 days' cooling method. Wagemann pointed out that carcasses frozen from four to six days, so that the internal temperature registered by spear-thermometers, reached -3°C., showed a loss in weight of 3.4 to 6 Kgm., which amounted to 204% to 2.43%. In comparison, Wagemann showed that in the case of 21 days' cooling, the loss of weight was 11 to 15 Kgms., amounting to 4.22% to 5.88%.

Wooldridge (1933) mentioned that when meat is frozen, the water within the muscle fibre separates out as ice, which is formed in the spaces between the fibres; on thawing, the separated water containing a certain amount of nutritive matter and haemoglobin, drains away. This "drip" is more copious with beef than with mutton. However, if beef is frozen sufficiently rapidly the ice is formed within the fibre and no "drip" occurs on thawing.

Keller advised that thawing should take place slowly, so as to avoid a

loss of juice. According to him a temperature of 5°C. is satisfactory. After the quarters have been thawed, the temperature should be quickly reduced by strong air current to about 0°C. This should be followed by a hanging in the halls for several days at a temperature between 0°C. and 4°C. to complete the "freshness" of the meat.

Kallert also suggested slow thawing of frozen carcasses, which should take four to five days, according to the weight of the carcass, at temperatures of 5°C. to 8°C.

At least one of the earliest investigators into the subject (Glage, 1896) pleaded that freezing was the most effective and safest methods of rendering measly pork fit for human consumption.

Reissmann (1897) found that <u>C. bovis</u> died within three days when kept at temperatures of -8°C. to -10°C. in the depths of pieces of meat.

Under similar conditions <u>C. cellulosae</u> died within 4 days.

Boccalari (1903) found that both <u>cysticerci</u> died within six days at 0°C. to -2°C. (Mönnig, 1928).

Ransom (1914) used two heavily infested bovine carcasses, which, after having been kept in a cooler for 24 hours, were quartered and hung in a freezing chamber at temperatures between 11°F. and 15°F. He retained one quarter in the chilling room as a control. After six days' continuous freezing he examined 63 cysticerci on a warm stage and found no movement. He then swallowed six cysts and after 18 weeks was still free from tapeworm infection. On the eighth day all twelve measles from his control quarter of beef in the chilling room were still alive.

Killisch (1923) found that by freezing half-carcasses of pigs at temperatures of -80°C. to -12°C. for 3½ days all cysticerci were dead. He performed his tests with five heavily infested pig carcasses, which

were delivered to him without skin or fat. He found that a 24 pound ham was frozen through after 48 hours, at a temperature of -8° C. to -10° C, and all measles were dead. In a 10 lb. ham, freezing was complete in the deepest muscles after 66 hours and all measles were dead after 76 hours at -6° C. to -8° C. In a $8\frac{1}{2}$ lb. ham freezing at -2.5° C. to -6° C. killed all measles in 125 hours. Similarly, at that temperature all measles were killed in a 20 pound ham in 150 hours. In a very large ham all measles were killed at -12° C. to -18° C. in 60 hours and with certainty within 72 hours at -10° C. to -15° C. He noticed that the circumference of a ham which measured 64 cm. around its widest part was increased by 2.5 cm. by freezing at -12° C. to -18° C. for 66 hours. Working also on C.cellulosae Brohmann (1924) confirmed Killisch's findings.

M.Muller (1923) confirmed the experiments of Wagner (1922) and showed that after 8 days' freezing of measly bovine carcasses no measles were found to be viable. He also tested <u>C.cellulosae</u> from measly pork carcasses which arrived at his abattoir in a frozen state. Some of these <u>cysticerci</u> were still able to show slight movements, but none evaginated their scolices. Muller names this as proof of the weak resistance of <u>C.cellulosae</u> to temperatures below zero.

Van Oijen (1929) and later van der Slooten (1936) pointed out the

"The only safe way is to freeze measly carcasses for at least 10 days at -10°C." (van der Slooten).

futility of treating measly carcasses under conditions of cooling.

Schmey and Bugge (1930 and 1931) found that isolated measles were killed in three to four hours by freezing at -8° C. to -10° C. They found that a temperature of -6° C. was reached in three to four days

in the innermost tissues of large pieces of meat at room temperatures of -8°C. to -10°C. According to those authors a freezing for 4 days is in every respect sufficient. Whatever refers to Chovis is equally applicable to C.cellulosae in pig carcasses. Lightly infected pork need thus not be boiled, but can be made safe for human consumption in four days by freezing at low temperatures. (Compare the results of our Bloemfontein investigations in section 6 of this Part.)

Von Ostertag (1930) once more advocated against the freezing of measly meat, and against Schmey and Bugge's recommendations in particular, i.e. freezing measly meat for three to four days at -8°C. to -10°C. His reasons were:- (1) Because after 21 days' chilling of meat, the measles evaginated their scolices, it did not follow that they were infective to man. (2) Freezing degenerated the meat, and there were not freezing chambers in all abattoirs in Germany.

Clarenburg (1932) found that in pieces of meat 6 cm. in thickness, which were placed in a freezing chamber at -8 °C. to -10 °C., the cysticerci were killed after 65 hours. He found that it took about 15 hours before the interior of the meat was cooled down to the same temperature as the air inside the chamber.

Feldforth (1934), in a series of experiments, exposed portions of beef infested with <u>C.bovis</u> to a temperature of -2°C. for varying periods of time. Viability was tested either by immersion of the cysts in a bile-saline solution at 41°C., or by swallowing the cysts in small silk bags. (Iwanizky's method). He found that 2 days at -2°C. were lethal to cysts in meat up to half-a-pound in weight.

Scheerer (1935) found that C. bovis rolled up in slices of meat and

exposed to a temperature of -2°C. are killed after 7 days and are no longer infective after 6 days. He concluded that slightly infested meat, frozen so that the innermost parts of the carcass remain at -2°C. for 6 days might safely be offered for sale. Zunker (1935) froze beef carcasses at an average temperature of -7°C. to -8°C. He read the internal temperatures of the meat by spear-thermometers, instruments encased in steel tubes with sharp points, which were inserted deeply into the musculature of the hind quarters. (Keller, 1935, in Zeitschrift für Fleisch-und Milchhygiene 45(17), pp. 321-322, had mentioned that in freezing cysticercous meat by the new rapid process, which aimed at maintaining the meat at -3°C. for 24 hours, it was not sufficient to control the temperature of the refrigerator. Keller mentioned that the more fat in a given carcass, the longer it would take to cool to -3°C. in the interior. Hence it was considered by him essential to read the temperature within the carcass. Keller described the use of a pointed steel tube, which was driven into the meat and which protected the glass stem-graduated thermometer.) Zunker explained that the kinds of changes which the animal tissue undergoes during freezing depends mainly on the speed of the freezing process. The faster the cooling down takes place, the finer is the crystal conglomerate in the frozen tissue. The killing off of living tissue depends on the complete freezing out of the fluid contents of, e.g. the bladderworm, and the speed with which this is done is an important factor. "Just as with special care living fish may be frozen and later again be thawed without suffering harm, so also the measles under advantageous circumstances can sometimes stand the freezing very well. During the slow freezing of the whole animal carcass, the measles are killed with certainty, however. "

Zunker found that it was necessary to keep an infected carcass in the freezing chamber until the temperature in the depths of the hind quarters registered -3°C. Even in the case of heavy carcasses, this took place within 7 days, and for safety's sake a further 24 hours could be allowed. In other words, Zunker recommended that measly carcasses should be frozen 24 hours longer than the time required for the temperatures in the deep muscles to reach -3°C. As an example he mentioned one quarter of beef, in which an internal temperature of -3.7°C. was reached in 5 days. That quarter, according to Zunker, was fit for issue on the 6th day. He was a staunch advocate of the freezing process of treatment for measly carcasses, in preference to the somewhat uncertain cooling process.

Kallert (1931) maintained that whereas certain plants and animals, according to Pictet, could withstand tremendous freezing, e.g., fishes frozen into blocks of ice at -15 °C. survived by careful thawing, but were killed outright at -20°C.; frogs could withstand a temperature of -28°C, millipedes -50°C., snails -120°C, cysticerci belonged to the creatures which were very susceptible to freezing and were definitely killed in freezing temperatures. He found that only at temperatures of -6°C. to -8°C. all meat juice was frozen. Kallert wuotes the experiments of Kallert and Plank, who found that hindquarters weighing 60 Kgm. were completely frozen through at temperatures of -6°C. to -8°C. in 6 to 7 days, while in fore-quarters this occurred in 5 days. According to Kallert, hind-quarters should be frozen at -8°C to -10°C. for at least 10 days, and fore-quarters for at least 9 days. He found that a superficial layer of fat greatly retarded cooling in a carcass, the heat conductivity of fat being 35% less than that of muscle. Large and heavy quarters required 3% longer time for complete through freezing.

Annie Porter, working in Johannesburg in 1923, "under commercial conditions", was probably alone among all modern observers in the results she obtained. She compared the resistance of cysticerci to the influence of cold, with those of fishes and snails, as was mentioned by Pictet, and felt that only after extremely prolonged freezing were cysticerci rendered innocuous. According to Porter's staining reactions, Chovis would appear normal in the deeper parts of the carcass after the 49th day of freezing, while some C.cellulosae appeared unchanged in the deep parts on the 156th day. Her freezing room temperatures varied between -7.28°C. and -16.2°C. On two occasions she swallowed C. bovis which had been frozen for 10 weeks and 12 weeks, respectively, with negative results. She also obtained negative results by feeding fresh and frozen Cysticerci cellulosae and Cysticerci bovis to puppies and rats. On the other hand, she based her criteria of viability of C. bovis and C. cellulosae under freezing conditions on several results she obtained from freezing C. fasciolaris and Echinococcus cysts, and thereby infecting a cat and a dog respectively. A rat liver, containing C.fasciolaris frozen for 30 days, and fed to a cat, which in due course developed T. taeniaeformis. A dog fed on a sheep lung containing Echinococcus cysts, which had been frozen for a month, developed T. echinococcus. Porter concluded that freezing at temperatures ranging from -5°C. to -18°C. for a period of about 10 weeks appeared to destroy the vitality of all the cysticerci in carcasses of beef and pork. For safety, according to Porter, a margin should be allowed on this, and a period of at least 12 weeks' freezing of slightly infested beef or pork at a temperature of 14°F., that is -10°C., should be undergone before the meat may be regarded as sterile and the cysticerci as dead.

A word of warning was expressed by Keller (1936), who stressed the necessity of maintaining a low temperature in freezing measly carcasses. He showed that by keeping pieces of beef infected with <u>C.</u>

bovis at temperatures between -1°C. and -1.5°C. not all cysticerci were killed even after 23 days at this temperature, but 50% were killed after 11 days. At such a temperature the meat is frozen, but the host capsule surrounding the cysticercus itself is not frozen. According to Keller, at such high temperatures of freezing, infectivity, tested by Iwanizky's method, is retained until at least the eleventh day.

According to Monnig (1928), about 1914, Ottesen found a method of

freezing fish in a 21 per cent. solution of sodium chloride at -10°C. to -15 °C., in which the fish froze ten to twenty times more rapidly and with much less loss of weight than in ait. Plank and Kallert (1915), referred to by Kallert (1923) and by Monnig (1928), confirmed the claims of Ottesen, but they found that with larger pieces of beef, half pig carcasses and whole sheep carcasses, the period of total freezing was shortened only about eight times. In the experiments described by Kallert, the temperature of the salt solution was -14°C. to -15°C. He pointed out that halves of pig carcasses which were frozen through in three days at -6°C. to -8°C.in the air, were frozen through in three to four hours in brine. claimed also, that smaller pieces of beef and lighter sheep carcasses would freeze sooner in brine. He found that there was less loss of weight of carcasses frozen in brine than of those frozen in air. According to de Jong (1922), meat frozen in brine left no salty taste, and could be delivered to consumers as in the case of fresh meat.

According to Kallert a small amount of salt did penetrate into the tissues.

Drooglever Fortuyn (1922) (Monnig, 1928) made comparative histological studies of normal meat, meat frozen in air, and meat frozen according to the above method. He found that refrigeration in air compressed the muscle fibres and drove them together in groups, relatively large cavities coming into existence between such groups; refrigeration in salt solution causes the appearance of cavities in the individual fibres but no compression of fibres.

Monnig (1928) mentioned that the <u>cysticercus</u> is well protected by its vesicle, and he was inclined to think that the more rapid death of the <u>cysticerci</u> under this method of treatment, as was found by Brohmann (1924). was due to the very rapid freezing.

According to Brohmann, Ottesen found more rapid freezing of fish in 21% brine, if the solution was continuously stirred. Brohmann did not stir the solution. He strongly recommended freezing measly pigs in brine at -15°C. He found that by freezing pigs in brine at that low temperature all measles were destroyed in 12 hours. Unfortunately, he made no infection tests, but relied mainly on evagination tests in bile-saline solutions, for his criteria. Monnig (1928) supplies the following summary of Brohmann's results:-

Shoulder	6	hours	in	brine	at	-6° to -8°C.	55/55	cysticerci	alive.
Ham	6	11	##	11	##	-10°C.	46/46	11	#1
Shoulder	8	##	##	11	##	-555°C.	40/40	91	11
	12	#	#	f 1	##	-10°C to -11°C.	42/42	11	#1
Shoulder	8	**	fi	Ħ	#1	-13°C.	403/40	11	11
	12	Ħ	\$1	11	#	-15 to -16.5°C.	0/48	Ħ	f1

"The pieces were all completely frozen when removed from the brine.

Cysticerci from all depths were examined after the pieces had been allowed to thaw at room-temperature." (Monnig)

Schmey and Bugge (1930) pointed out that refrigeration technique had

advanced so far that refrigeration in brine would result in obtaining a temperature of -6°C. in a few hours.

CE

VIABILITY TESTS WITH MEASLES TAKEN FROM CHILLED AND FROZEN PORK AND BEEF CARCASSES AT THE BLOEMFONTEIN ABATTOIR.

The object of these tests was mainly to confirm the tests of overseas writers, and, if possible, to establish definitely, with what material we chad available, the shortest period required for the freezing of measly beef and pork carcasses at temperatures of approximately -10°C., under South African commercial conditions.

Method Employed.

- (1) After the stipulated period of chilling or freezing, the cysticerci (cellulosae or bovis) were always removed from their connective tissue capsule.
- (2) Physical characteristics of such treated <u>cysticerci</u> were observed, although no microscopic observations of loosening of the hooks in C.cellulosae were made.
- (3) Control evagination tests in bile-saline solution and in sodium taurocholate were made in an incubator at 38 °C.
- (4) The main criteria in these tests were based on actual infection tests on several voluntary assistants, according to both Iwanizky's and Keller's methods.
- (5) In view of the fact that the infection tests perfected by Iwanizky and by Keller have very nearly approached conditions of natural infection, it was not considered necessary to attempt reactions to warming on a stage, and the observations of the physical characteristics of

frozen or chilled measles were merely those of noting cloudiness, discolouration and consistency of the fluid.

(6) Since the boiling or pickling of measly carcasses is not practised in South Africa, such tests were not attempted. All available material was, therefore, used in cooling and freezing tests.

With regard to control artificial evagination tests, we found the best results with 5% sodium taurocholate solution (Malkani's method). The cysticerci were carefully isolated from their connective tissue capsules and placed in a saucer containing 5% sodium taurocholate solution, which was then placed in an ordinary "Buck-Eye" egg incubator, of which the heat was regulated to 38°C. Evagination of scolices usually occurred within two hours in the case of viable cysticerci. It was found that this method worked very successfully with fresh (unfrozen) cysticerci, more sluggishly with those frozen for about 24 to 48 hours, or chilled for about 21 days, and frequently did not work with those measles from meat chilled longer than about 21 days, or frozen longer than 48 hours. On the other hand, we established definitely that the only sure criterion of viability of measles could be obtained by actual infection tests according to Iwanizky's and or Keller's methods. In some cases we failed to obtain evagination in vitro of scolices of measles from meat frozen for three days and more, whereas ready and clear evaginations of such scolices occurred within the bag or tube in our subject's intestine. We used a slight modification of both Iwanizky's and Keller's methods. In the former we found that frequently took some time to stitch the bags, as Iwanizky did, and frequently the sutures did not appear too secure. In order, therefore, to instil the fullest confidence in his personal safety

in our subject, we tied the small silk bags with strong suturing silk, and cut the tied end as short as possible. All our subjects, European members of the Abattoir Staff, had no difficulty in swallowing them, and one subject in particular, never failed to recover the bags, or the celluloid tubes in his stools. With reference to our modification of Keller's method, we found the same objection to suturing the silk cover. A single layer of silk was, therefore, wrapped round the celluloid tube containing from one to three measles, in such a way that the two open ends of the small cylinder were covered with a single layer of silk, drawn tautly, and the four corners of the silk coverlet were twisted together in a spiral and tied as closely to the body of the cylinder as possible. This spiral twisting caused the two single silk-layers covering the ends of the tubes to be drawn even more tightly, almost like a drum, then in the case of suturing the coverlet, or else fixing by means of artificial fixatives, e.g. acetone-cellulosa solution, which took a considerable time to fix, and was treated with a certain amount of suspicion by our "chief" subject. Most of our subjects experienced a measure of discomfort in swallowing the hard celluloid tubes, measuring approximately 15 mm. long by 7 mm. diameter. Our "chief" subject, however, once more found no difficulty in the deglutition of the somewhat unwieldy "pills", and for that reason experiments by means of Keller's method were confined to him, since he was quite prepared to use the celluloid tubes over and over again, he never "lost" any, and I was only able to obtain the tubes through the kind favour of the Director of Veterinary Services, Onderstepoort Laboratory, and could therefore, not abuse his favour by repeatedly applying for fresh tubes. The inner temperature of the meat in the freezing chamber was read on improvised spear-thermometers, ordinary low graded freezing thermometers, encased in sharp-pointed steel covers. One of these was kindly made for me by the Mechanical Engineer in charge of my Abattoir Refrigeration plant, and two were lent by the Director of Veterinary Services, Onderstepoort.

TABLE A.

Evagination Tests with C.cellulosae - Chilling Tests.

ExPERIMENT	PART OF CARCASS	(WEIGHT)	No DAYS CHILLED	METHOD USED Istuanizhy K, Keller	No. of Human	Subfects	No. of Tubes or	BAGS SWALLOWED	NUMBER	RECOVERED	TOTAL NO. OF CYSTS SWALLOWED	No. 05 50011- CES EVAGENA- TED	NO. OF CYSTS DIGESTED (DEAD)	REMARKS.
1	(21	llder	4	I.		3	10	В,		9	10		3	l bag was lost; 6/10 scolices evaginated in 5% Sod. taurocholate sol.
2.		ılder lbs)	6	I.		2	4	В.		4	8	6	2	4/10 scolices evagin. in 5 % sod.taurochol.
3.	Leg (18	lba)	1	K.	· · · · · · · · · · · · · · · · · · ·	1	4	T.	· ·	4	8	8	0	30% pig bile.
4.		lbs)	. 2	K.		l	4	T.		4	8	7	1	4/10 scolices evagin. in www.www.www.www. 30% pig bile.
5.	Leg (27	lbs)	3	., K.		1	3	T.	1	3	9	9	0	
6.	Leg	lbs)	4	К.		1	3	T.		3	6	6	0	6/10 scolices evagin. in 5% sod. taurochol. & 4/10 in 30% pig bile.
7.	Leg		5	K.		1	2	T.		2	6	6	0	
8.	Shor	lder lbs)	6	K.	;	2	2 2 2	T. B.		4	!	10	2	5/10 scolices evagin. in 5% sod. taurochol.& 2/10 in 30% pig bile.
9.		lder lbs)	7	K. I.		2	2 3	T. B.		5	14	13	1	3/7 scolices evagin. in 5% sod. taurochol.& 1/7 in 30% pib bile.
10,		ilder lbs)	8	K.		2	3 3	T. B.		5	17	14	0	3 cysts in 1 bag lost.
11.	Leg (-)		9	K I.		1	3 3	T. B.		6		16	0	2/5 scolices evagin. in 5% sod. taurochol. & 1/6 in 30% bile.
12.	Leg		11	K.		ı	3 3	T. B.		6	16	16	0	
	C11- a.	. 7	10	K.		1	, 3	Т. В.				<u> </u>		
13. 14	Leg	lbs)	,	I.	J	1		В,	erent a n	<u>6</u> 5	12 15	<u>~</u>	<u>3</u> 15	Meat was badly putre- fied, and measles were stickily opaque. All artificial tests faile
15.	Leg (32	lbs).	14	K.		1		T. B.		6	18	10	8	
16.	Leg (30	lbs)		K. I.		1.	2 2	т. В.		4	10		0	5/6 scolices evagin.in 5% sod. taurochol. & 2/7 in 30% pig bile.
17		lder lbs)	16	K. I.		1		Т. В.		4	10	10	0_	
17.	Neck			I.		<u> </u>		В.		2	4			
18	(10	T/00 /										_		iversity of Pretoria, Library Services, 2013

TABLE A2 (continued).

ExPERIMENT IYUMBER	PART OF CARCASS	(WEIGHT.)	NO OF UAYS CHILLED	METHODUSED J. IWANIZKY K. KELLER	HUMAN SUBSECTS	No OF TUBES OF BACS	SWALLOWED	NOMBER RECOVERED	FOTAL NO OF CYSTS SWALLOWED	No of Scources Erronnaried	NO OF CYSTS DIGESTED (DEAD)	REMARKS.
19.	Leg (21	lbs)	17	К.	_1	5	T.	5	15	7	8	: 1
20.	Leg	lbs)	18	K	1	4	T.	4	12	9	3	3/6 scolices evagin in 5% taurochol. & 0/5 in 30% pig bile.
21.	(17	lder lbs)	19	K. I.	2	6 4	-	10	30	29	1	
22.	Shou (19	lder 1bs)	20	K.	1	5	T.	5	15	13	2	ļ
23.	Leg (36	lbs)	21	K. I.	2	4 3	T. B.	6	20	15	2	l bag with 3 cysts lost; 3/10 scolic. evagin. in 5% Sod. taurochol. & 2/10 in 30% pig bile.
24.	Part Leg	of	22	К	1	4	T.	4	12	8	4	
25.		lbs.)	23	к.	1	4	T.	4	10	10	0	
26.	Part Leg		24	K,	1_1_	4	T.	4	10	10	0	
27& 28	Leg Shou		25	K.	1.	3 5	T. T.	8	25	6 13	4 2	Two experiments on subsequent days.
29& 34		lder å	1	к.	1.	5 4	T. T.	9	25	12	3 10	Two experiments with 10 days inter-
30& 35	Leg Leg	tuurooluuse Millerin Millerin Millerin een Alleri	27	К.	1.	5 5 5	T. T.	10	30	12	3 12	Two experiments & pork from same respective carcasses as 29 & 34.
31 & 36	Shou Leg	lder	28	K. I.	2		r.2B. r.3B.		39	7 9	8 15	Two experiments, pork from differ- ent carcasses
32 & 37	Leg Shou	lder	29	K.	2		r.2B r.4B		35	5 7	10 13	Two experiments, pork from same carcasses as 31&36
3 3 &	Leg Leg		30	K. I.	3	4	r.2B.		32	7	10 15	Two experiments with different carcasses.
<u>38</u>				K.		6.	T.					Two bags with 6
39. 40.	Shou Leg	Laer	31 32	K. I.	2	5 5 5	T. B.	10	30 31	13	18	No artificial tests tried with experi-
				K.	~~~		T.			·		ments 24 to 42 incl. 1 bag with 3 cysts
41.	Leg	-	33	I.	_1_	<u>5</u>	B. T.	9	30	3_	24	lost. 0/10 scolices evag.
42.	Shou	lder	34	I.	1	5		10	30	12	18	in 5% taurocholate a 0/9 in 30% pig bile
43.	Shou	lder	35	K. I.	1	5 5	T. B.	10	30	1	29	do.
44.	Leg		36	I.	1	3	В,	8	24	0	24	do. 2 scolices in 2
45.	Leg		37	K. I.	1.	4		8	24	2	22	tubes evaginated, none in the bags.
46.	Carc	ass	38	K. I.	1	4		8	20	0	20	and the second s
47.	Carc	ass	39	K. I.	1	4	T.B.	_5_	14	0	14	and the same of the same and the same
48	Carc	ass	40	K. I.	1	5 6	T. B.	8	40	0	32	3 bags lost with &
49.	Leg		41	к. 1.	1		Т. В.	5	15	1.	14	
50.	Leg		42	K. I.	1	3 2	T. B.	5	15_	0 0	15	
50.												

Evagination tests with <u>C.cellulosae</u> from pork cooled for various periods have thus shown that the most successful results have been obtained with natural infection tests, according to Iwanizky's and/or Keller's methods,

In five per-cent. sodium taurocholate, evagination nearly always occurred within two hours, and by using 30 per-cent. pig bilephysiological saline solution in from 2 to 5 hours. After that. evaporation of the fluid contents of the saucer often occurred. fresh measles, namely those which had not undergone prolonged chilling or freezing, evagination occurred more readily, and in a shorter time. After about the twenty-first day of chilling, putrefaction frequently set in, in the pork, but, according to the results obtained by our natural infection tests, it did not follow that the measles situated in badly putrefied areas, died in that situation. By our tests we established the fact that the Cysticercus cellulosae can remain infective up to 41 days after slaughter of the host, but we failed to find any alive after that day, although we only tried two more tests, on account of the undesirability of maintaining putrefied neat in the condemned meat section of our chilling room.

It is interesting to mention that after 35 to 41 days' chilling, we found only 4 out of 167 tested measles viable. After 34 Tays' chilling, 12 out of 30 pig measles were still viable; and after 30 to 33 days' chilling, 43 out of 123 pig measles tested were till viable.

For illustrations see Figures 3 to 6.



Fig. 3.

C.cellulosae scolices evaginated in 2 hours in 30% pig bile-saline solution. (2 days chilled) Magnification 7 times.



Fig. 5 a.

C. cellulosae scolices evaginated Keller's method after 28 days' cooling. Magnification 7 times.



Fig. 4.

C.cellulosae scolices evaginated by Keller's method, after 19 days' cooling. Magn. 7 times.



Fig. 5 b.

Microscopic view of head of same, showing suckers and rostellum.
Magnification 40 times.

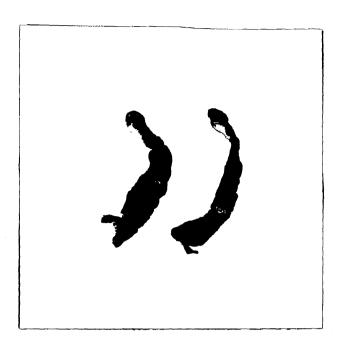


Fig. 6.

C. cellulosae scolices, evaginated Keller's method after 32 days' chilling.
Magnification 7 times

TABLE B.
Freezing Tests with Cysticercus cellulosae.

-	سياسياس								0100	LUUS	0 (3.1	444	osae.						
	NO OF DAYS IN FREEZER	TEMP. OF MEAT AFTER 24 HOURS COOLING .F.	FREEZING CHAMBER OF	TEMP. OF FREEZING AFTER	W) TEMP. OF MEAT 2) TEMP. OF FREEZER OF. AFTER 24 HOURS	(1) TEMP. OF MEAT OF.	AFTER 48 HOURS	W TEMP. OF MERT OF. W TEMP. OF FREEZER OF.	W TEMP. OF MEAT OF. 18) TEMP. OF FREEZER OF.	(1) TEMP. OF MERY OF. (2) TEMP. OF FREEZER OF.		AFTER SIR OAYS	W TEMP, OF MEAT P. F. (2) TEMP. OF FREEZER OF PFER SEVEN DAYS		(WEIGHT)	METHOD IMMNIZKY I	SWALLOWED	DIGESTED (DEAD)	Kemarks
1	14			-	timiti al film and a second	-									Car- cass		35	_35,	1
2	12								-						Car- cass	- Consideration - Affile (c.)	25	25	2
3	6		13	The second secon	1) 2)14	1,2)	13	1) -	1))	- 1) 4 2)	- 1 12 2)13	-		Leg 30 los	I.	12	12	8
4	. 4	i	12		2)13	1)	13	2)12	2)1	6	i		P. A. Marrielle, and Quantity on		Shou.	I K.	4	4	6 _
5	1		15	-	2)14	1		And the second s							Leg 241b	K.	6	6	3
6	2	-	14		1)19 2)13	1)	14			Ž	· ·	pum MethaMac AM	re rije aartik op filjen entgeret		Leg 261b	K	6	3 3	4
7	3	:	17		1)18 2)12	2 2)	18 14	1)17 2)15			***				Shou:	i K	10	7 3	5
8	4				2)15	(2)	17 16	1)17 2)13	$\begin{pmatrix} 1 \\ 2 \\ 1 \end{pmatrix}$	4				is the second	do. 15 1	þ K	12	1 11	6
9	5	40	1		2)15		17 16	2)13 1)17 2)13	1)1 2)1	42)					161b	I	8	8	7
10	6	40	• .	1	do		do	do			do 1 2	15 15 do	-	and art	Le g _331b	K	18	18	8
11	7		do		do		do	do		-	do	do	1)16 2)18	1	381b Leg	K	13	13	9
12	4	41	18	19	1)18 2)15	1)	18 13	1)16 2)11						đ	houl- er 16	I	18	18	6
13	5		Ì.	do			do.	d.o	a d	$\left. egin{array}{c} 0.1 \ 2 \end{array} ight\}$	18 14				o. 91bs	K	18	18	7
14	1	39	15	15	1)20 2)16					. Section (Section)				I	eg 11bs	KI	15	5 10	3
15	2	do	do	do	1)20	1)	18 15					pentilipe collis is til			e g 91bs	K	15	15	4
16	3	do	do	do			do	1)18 2)15				erer den p lantal	28-1815 1817 1824 1 Base	S	houl- er 18	K	10	10	5
17	4	ďο	do	do	do)	do	do	1)1				-	****	do. 9 1b	K	15	15	6
18	1	41	19	119			-		~ / _ '			Principal const		Ī	eg 2 1b	K	7		3
19	2	dо	do	dо	2)16 do	1)	20							Ī	eg 41b	K	8	8	4
20	3	do	đo	do	đơ		15 do	1)18		Confidence Management of the	non face describe of t	P. (2000)	nin Yardi dan melammaga, d	Ī	eg 6 1b	K	8	8	5
21	3	40	20	20	1 23			2)16				-		C	arcass 85 lbs	зK	60 4		5
22	4	36	14	,16	2)17	1)	21		1)18						do. 87 lb.	K	40	40	6
23	4	38	16	16		1)	17	1)17		5	-	grama _{a -}			do. 06 1b	K	36	36	6
24	5	40	13	14	2)14 1)19	1)	18	2)1 <u>1</u> 1)18	$\frac{2}{1}$	3,1)	17		Section for providence		do. 71 1b.	K	50	50	7
25	5	41	15	16	2)16 1)23			2)15	2)1;	3 2). 7 1).					do. 61 1b.	K	40	40	7.
					2)16				2)1		9	igitise	d by the	-	rsity of Pre				s, 2013

TABLE B. (continued).

ELPER MENT NUMBER	NO OF UAYS IN FREEZER	THE OF MENT AFTER THE	MITTER TEMP. OF FREEZING CHAMBER F.	11	10 TEMP OF PREEZER OF	HETER 24 HOURS	W) Temp of Pacetin of		W TEMP OF FREEZER OF	AFTER THREE DAYS	(3) TEMP OF MERT OF	AFTER FOUR DAYS	(1) TEMP OF MEGT OF	AFTER FIVE DAYS	TEMP OF MEAT	W TEMP OF FREEZER OF	HFTER SIN DAYS	O TEMP OF MEGT OF	AFTER SEVEN DAYS	PART OF GARCASS	(WEIGHT)	,	I WANIZKY	001	NO OF CYSTS	SCOLICES EVACINATED	DIGESTED (DEAD)	REMGAKS.
26	5	38	12	13	1)	20 11	1)1 2)1	.6	1):	16 14	1 2)15)11	1 2)15)12			-				Car	cas 1b	8	K	30	0.	-30	7
27	5	39	18	16	1)	18 14	1)1 2)1	.8 .5	1):	17 13	1)	14	1)15)13			;				do 1	1b.	:	K I	40	0	.́40	7
28	5	40	16	15	1) 2	17)15	1)1 2)	.6 142	1): 2)1	16 4	1) 2)	15 13	1 2))14 13					1	٤	do 30	lb.		K			30	
29	5	39	16	16	1)	21 14	1)2 2)1	0 5	1)] 2)]	19	1)	18	1 2)17)13			:			10	do 1	lb.		K I	30	0	30	7 yanuda
30	5	41	14	13	1)	20 14	1)1 2)1	.8 3	1)] 2)]	L7 L3	2)	17 15	2)17)17				-		. 6	do.	lb.	:	K. I	60	0	60	7
31	5	39	13	14	1):	18 13	1)1 2)1	6	1)] 2)]	l.6 l.4	1) 2)	15 13	1 2)15)15							do 1	lb.		K. I	35	0	35	7
32	5	42	14	14	1);	22 15	1)2 2)1	2 3	1)2 2)1	21	1)2)	20 16	2)16)12				-		20	do.			K.	20	0	20	7
33	5	37	18	17	1)	19 17	1)1 2)1	8 5	1)1 2)1	18 17	1)	18 12	1,2	17			-	No. of Species	•••••		do.			K. I	30	0	30	7
34	5	38	17	17	1)2	21 : 17 :	1)2 2)1	1 6	1)2 2)1	14	1 }	20 15	1 2)19)13			:	-	-		do. 8			K.	30	0	30	7
35	5	39	16	18	1):	20 16	1)2 2)1	1 5	1)2 2)1	17	1)	18 14	1 2)18)12				- Company	-		do.			K I	30	0	30	7
36	5	40	15	16							:					-			****	13	do.			K.	24	0	24	7
		41									1		1		1			و من المنواد			d o.		:	K.		0	30	7
		1	18		1			- 1			\$				į						do 6	l b.		K I	30	0	30	7

Remarks Index to Foregoing Table (B).

- 1. A heavily infested pig carcass was frozen for 14 days. The weight of the carcass was not recorded at the time. The carcass was literally frozen to resemble a wooden box in consistency. Some of the <u>cysticerci</u> were removed and resembled crystals of ice. The carcass was thawed for 24 hours and 35 measles were removed from the innermost muscles. These were swallowed in their naked state by five members of the Abattoir Staff, N.F.V.; H.M.D.; MC; P.J.K.; and W.H.G.; each of whom swallowed seven measles. After twelve months, none of the subjects has developed tapeworm infection.
- 2. A heavily infested pig carcass was frozen for 12 days. Its physical condition resembled that of Experiment 1, after freezigq. The carcass was thawed for 24 hours and 25 measles were removed from the deeper tissues and swallowed in their naked state by two natives on the staff and one European, R.P. After approximately 12 months, no tapeworm infestation has resulted.
- 3. Experiment No5: Leg of Pork weighing 24 lbs. was frozen for 24 hours. The leg was practically frozen through. Six cysticerci were swallowed by W. H. G. according to Keller's method, and six scolices evaginated.

Experiment No. 14:- Leg of Pork weighing 21 lbs. was frozen for 24 hours. Fifteen cysts were swallowed by W.H.G. in 2 tubes and 3 silk bags. All were recovered. Five scolices (2 in each tube and 1 in 1 bag) had evaginated, and 10 presumably dead cysticerci were completely digested.

Experiment No. 18: - Leg of Pork weighing 42 lbs. was frozen for

24 hours. This was a fairly fat animal. Seven <u>cysticerci</u> in two tubes were swallowed by W.H.G. Both tubes were recovered. One contained 3 evaginated scolices and the other, one. Three <u>cysticerci</u>, presumably dead, were digested.

Contemporary artificial evagination tests were made, with the following results:-

2 out of 6 scolices evaginated in 5% sodium taurocholate solution.

1 " " 5 " " 30% pig bile-physiological saline (solution.

On three occasions W. H. G. swallowed a total of 28 <u>C. cellulosae</u>, which had been removed from pork frozen for 24 hours. Out of this number 15 scolices evaginated.

4. Experiment No. 6:- Leg of Pork weighing 26 lbs. was frozen for 48 hours. Meat was nearly frozen through. After 24 hours' thawing of the meat, W.H.G. swallowed two celluloid tubes, each with three measles. A total of three out of the six measles evaginated scolices. (This leg of pork was from the same carcass as that of Experiment No. 5, above.)

Experiment No. 15:- Leg of Pork, 19 lbs., was frozen for 48 hours. Freezing, in this case, was definitely complete. After 24 hours' thawing, 15 measles were removed and given to W.H.G. to swallow in 3 tubes and 2 bags. No scolices evaginated, and all the measles, presumably dead, were digested totally. (This leg of pork was from the same carcass as that of Experiment No. 14, above.)

Experiment No. 19:- Leg of pork weighing 44 lbs. was frozen for 48 hours. Fat leg. Leg was nearly frozen through. After 24 hours! thawing of the meat, W.H.G. swallowed 8 measles in three tubes. All were digested. Artificial evagination tests in 5% sodium

taurocholate and 30% bile-saline solution were equally negative.

This leg of pork was from the same carcass as that of Experiment 18.

Summary: On three occasions W.H.G. swallowed 29 measles from pork frozen for 48 hours. Of this number, only 3 scolices evaginated.

Experiment No. 7:- A shoulder of pork weighing 19 lbs. was frozen for three days. The shoulder was completely frozen through, and many of the measles resembled ice crystals. After 24 hours (thawing of the meat, W.H.G. swallowed 10 measles in three tubes. The cysts were removed from the deeper part of the subscapular muscle. Of this number, 7 scolices evaginated, and three cysts were digested. The other shoulder and both hind legs were used in cooling tests. (See chilling tests Nos. 15 to 17, which showed fairly regular evagination results.)

Contemporary artificial evagination tests revealed the following negative results: 0/6 scolices evaginated in 5% sodium taurocholate solution; 0/7 scolices evaginated in 30% pig bile-saline solution.

Experiment No. 16: Shoulder of pork weighing 18 lbs. was frozen for 3 days. Freezing was complete. After 24 hours thawing of meat, W.H.G. swallowed 10 measles in 3 tubes. All were digested.

Experiment No. 20: Leg of Pork weighing 26 lbs. was frozen for 3 days. Freezing was complete. After 24 hours thawing of the meat, W.H.G. swallowed 8 measles. All were digested.

Experiment No.21: - A whole carcass weighing 185 lbs. was frozen for 3 days. Pig was of the large-boned, heavy variety, but not very fat. Freezing was quite complete. After 24 hours' thawing of meat, 60 measles in 5 tubes and 15 bags were swallowed by W.H.G. and three others. Two of the assistants "lost" their bags.

No scolices evaginated.

Summary: On 4 occasions W.H.G. (assisted by three other persons on the last occasion) swallowed a total of 88 measles from pork frozen for three days. Of this number, 7 scolices (all on the first occasion) evaginated.

Experiment No. 4:- Shoulder of pork weighing 18 lbs was frozen for 4 days. Freezing was complete. After 24 hours' thawing of the meat, W.H.G. swallowed 4 cysts in 2 tubes. All measles were digested.

Experiment No. 8:- Shoulder of pork weighing 15 lbs. was frozen for 4 days. Freezing was complete. After 24 hours' thawing of the meat, W.HG. swallowed 12 measles in 4 tubes. Of this number, 1 scolex evaginated.

Experiment No.12:- Shoulder of pork weighing 16 lbs. was frozen for 4 days. Freezing was complete. After 24 hours thawing of the meat, W.H.G. swallowed 18 cysts in 6 silk bags. All measles were totally digested.

Experiment No. 17:- Shoulder of pork weighing 19 lbs. was frozen for 4 days. Freezing was complete. After 24 hours' thawing of the meat, W.H.G. swallowed 15 measles in 3 tubes and 2 bags. All measles were totally digested.

Experiment No. 22:- Whole carcass weighing 87 lbs. was frozen for 4 days. Freezing was complete. After 24 hours thawing, W.H.G. and P.J.K. swallowed 40 measles in 5 tubes and 8 bags. All receptacles were recovered. All measles were digested.

Experiment No. 23:- Whole carcass weighing 106 lbs. frozen.

W.H.G. and P.J.K. swallowed 36 measles. All measles were totally digested.

Summary: Out of a total of 125 measles obtained from pork frozen for four days, and swallowed by W.H.G. and an assistant, only one scolex evaginated.

7. Experiment No. 9:- A shoulder of pork weighing 16 lbs. was frozen for 5 days. Out of 8 measles swallowed, all were digested.

Experiments Nos. 24 to 38:- Fifteen whole carcasses of pork, weighing from 36 lbs. to 202 lbs. were frozen for 5 days. The heaviest carcasses, 161 lbs.and 202 lbs., were very fat. In each case the carcass was frozen right through. From these fifteen carcasses a total of 509 measles were swallowed by W.H.G. and three others. Of this number no sclblices evaginated. A few silk bags were "lost", but all measles in those recovered showed complete digestion.

From four of the various carcasses contemporary experiments were tried in 5% sodium taurocholate solution and in 30% pig bile-physiological saline solution at 38°C. incubator temperatures, but no scolices evaginated.

Summary: Out of 535 measles swallowed by my assistants, no scolices developed. The measles had been recovered from pork frozen for 5 days.

8 & 9 Experiments 3, 10 and 11:- Three legs of pork, each weighing approximately 30 lbs., were frozen - 2 for 6 days, and 1 for 7 days.

Out of 43 measles swallowed, none evaginated their scolices.

The following short table summarizes the results of our freezing tests with Cysticercus cellulosae.

Days frozen	Total Measles Swallowed	Total Evaginated	Digested or Lost	Percentage Viable.	***
1	28	15	13	53.57	
2	29	3	26	10.35	
3	88	7	81	7.95	

205. 265°.

(Table continued	.)	d	e	1	nı	Ĺ.	j	t	n	0	С	е	1	b	a	Ί	1
------------------	-----	---	---	---	----	----	---	---	---	---	---	---	---	---	---	---	---

ays frozen.	Total Measles Swallowed.	Total Evaginated	Digested or Lost	Percentage Viable.
4	125	1	124	0.80
5	535	0	535	*
6	30	0	30	•
7	13	0	13	-
12	25	0	25	*
14	3 5	0	35	₩

Conclusions.

From our experiments with the freezing of measly pork, it will be noticed that at freezing room temperatures ranging from 14°F. to 19°F. (-10°C. tc -7°C. approximately) and an internal temperature of the deeper tissues of a leg of pork of 20°F. to 23°F. (-7°C. to -5°C.), after 24 hours freezing. Cysticercus cellulosae is still viable.

The percentage of viable measles found steadily diminishes from 2 days' freezing to 4 days' freezing, until after 5 days' freezing no more viable Cysticerci cellulosae were found. In view of the fact that one viable measle was found after 4 days' freezing, it would be dangerous to recommend that pork frozen for 4 days at temperatures oscillating between 13°F. and 16°F. (-11°C. and -9°C.) would be safe.

According to Schmey and Bugge, four days' freezing is sufficient to kill all <u>Cysticerci cellulosae</u>, and according to Killisch, all <u>C.cellulosae</u> are destroyed in half-pig carcasses in 3½ days. At Bloemfontein, on the other hand, we found that from a light shoulder of pork, weighing only 15 lbs., and frozen for four days, 1 out of 12 measles was still viable. It is interesting to record that a temperature of 19°F. (-7°C.) was reached in the depths of the subscapular muscle within 24 hours, and of 16°F. (-10°C.) within 4 days.

From 17 experiments performed with measles from pork frozen for

five days (including 15 whole carcasses), our results justify the presumption that even in heavy and fat (202 lbs.) pig carcasses, <u>Cysticerci</u> cellulosae are destroyed. In the 202 lbs. pig carcass (Experiment No.32) the inner temperature registered by means of a steel-pointed thermometer inserted deeply into the musculature of the hind leg, reached 22oF. (-5.5°C.) in 24 hours, and 16°F. in 5 days. During that period the freezing room temperature oscillated between 12°F. and 16°F.

It is presumed that very few pig measles will remain viable if subjected to continuous freezing at -10°C. in pork, for 5 days. A safety margin of 2 days can be allowed, and after 7 days' freezing, lightly infested pig carcasses, provided they are not too fat, can safely be passed as fit for human consumption. There can be absolutely no objection, from a public health point of view, to the treatment of lightly infested measly pork carcasses, no matter how fat they are, for fourteen days at -10°C. continuous freezing, as South African Meat Regulations provide for at present, although few abattoirs make use of the concession.

Chilling Tests with Cysticercus bovis.

Only four such tests were performed at Bloemfontein, and one ox carcass was quartered, the quarters being kept in the cooler for 20 days, 27 days, 30 days and 31 days. Viability of the cysticerci, according to Keller's method, was tested, and scolices evaginated to the extent of 6 out of 14, from a fore quarter chilled for 20 days. No scolices evaginated from 17 measles from a hind-quarter chilled for 27 days. Putrefaction had, by then set in. At 30 days, the remaining fore-quarter and hind-quarter had badly putrefied, but, mevertheless, ten measles were removed from each after 30 days' and 31 days' chilling, respectively.

These were also tested according to Keller's method, but no evagination of scolices occurred.

Since South African Regulations do not prescribe a period of 21 days' chilling, or longer, as an alternative to the freezing method of rendering slightly infested measly beef fit for human consumption, I did not consider that any useful purpose was served by using our available material on chilling tests. Secondly, it was very undesirable to keep rapidly decomposing beef in the condemned meat section of the abattoir chilling rooms.

Our viability tests with beef measles were, therefore, mainly confined to freezing tests, the results of which are tabulated on the subjoined table.

As with <u>C.cellulosae</u>, we performed a number of contemporary tests in 5% sodium taurocholate solution, and in the case of <u>C.bovis</u>, with 30% ox bile-physiological saline solution, instead of pig bile. Viability of the fresh measles was always tested by Keller's method, prior to the carcass being placed in the freezing chamber. Twenty-four hours' cooling of the carcass was allowed in every case, prior to freezing.

206. 268

TABLE C.

									_1	AE	LE	<u>c.</u>													
		Free	ezi	ng	T	est	8 V	vi t	h C	ув	tio	cer	cus	3 t	ovi	<u>s</u> .									
NUMBER NO DE OGYS IN FREEZER TEMP. OF MERT AFTER ALHOURS COOLING OF	INITIAL TEMP. OF FREEZINGCHAMBER	TEMP. OF FREEZING CHAMBER AFTER S. HOURS OF.	(1) Temp. OF MEAT OF. (3) TEMP. OF FREEZER F	AFTER 24 HOURS	(1) TEMP. OF MENT OF.	AFTER WS HOURS	(1) TENP OF MERT OF B) TEMP OF FREEZER OF	AFFER 3 DAYS	4) TEMP OF MEAT OF	AFTER 4 UAYS	UTENID OF MEAT OF	AFTER S DAYS	W TEMP OF MEAT OF	AFTER 6 DAYS	() TEMP OF MEAT OF	AFTER 7 DAYS	PART OF CARCASS	(587)	METHOD USED	No of cysts Swallowed	FY OF CASTS	NO OF CYSTS DIGESTED (DEAD)		IN O.E.K.	
1 1 44	1			29 14							-			-			Side	e oi			4	0	1		
2 2 41	The second		1)2)	28 15	2)	27 13							Action and with the second			7	Sid bee: 262	e 01 f	K	2	0	2	2		
3 2	16			17	1) 2)	12							Process Comment		AND	1	0x Tong			8	0	8	2		
2 1 44	20		2)									-	madel and square a save		To the second se		361	beed 1b.		10	10	0	1		
5 3 44	20		2)	22 9	2	22 11	2)	20 10		-		**	1				S1d 350	1b.	K	14	7	7	3		
6. 2 41	12		2)	24 11	2	11							-	-		1	Side 254 Side	1b.	K	12	1	11	2		
7. 1 39	12		2)	23 15 25	71	23		23					4_		-		245 Side	lb.	K	8	8	0	1		
8. 3 41	14		2	16 28	2	14 24	2)	23 23	_			-		Dave gar	<u> </u>	to favorite	250 Side	1b.	K	12	9	3	3		
9. 3 42	14	_		18 28	2)	12 22	(2)	11	7 1	14			-		-	1	248 Side	lb.	K	10	2	8	3		
10 4 43	19	14	2)	ĩ0	2	12	2)	23 19	1 2	14	-	12	<u> </u>		-		300 Side	lb.	K.	10	4	6	4		······································
11 5 43	19	14	đ	0	11	<u>do</u>	77	10	7	<u>do</u>	2	iĩ					288 Hind	1b.	K	13	1	12	5	;	
12 4 42	21		77	21		23 16		19 11		20 15 19	<u>.</u>	18		17		and the second	Qtr 160 Side	1b.	K	10	4	6	4		
13 6 41	23	15	2)	21 18 21	2)	17 20	רדו	20 16 20		17 19	T	15	ຸ່ 2)	11 13	: :). 1	289 Side	1b.	K	12	0	12	6		
14 7 40	23	15		18 22	2)	17	2)	16	2)	17	7 -	15	2	11	2)	13	290 Side	1b.	K	9	0	9	7		····
<u>15 5 41</u>	20	18	2)	17 22	2)	16	2)	14	2)	15	2	12		16	-		250 Side	lb.	K.	11	0	11	5		
16 6 80	19	18	2)	<u>ĩ</u> ỡ	2)	<u>15</u>	2)	13	2)	13	2	12	2	13			316 Side	1b.	K	15	0	15	6	-	
17 7 do	đo	do	<u>d</u>	0 21	77	<u>do</u>	1	<u>do</u>		<u>do</u>		do	-	do	2)	15	312 Side	1b.	K	12	0	12	7		
18 4 42	13	15										17	-		 		281 Side	1b.	K	7	2	5	4		
19 5 do	do	do	11	<u>do</u> 25		24		23			42	14	-		 		289 Car	1b.		11	0	11	5		
20 5 41	16	16	2)	15	2)	13	2)	13	2)	11	2	12	-		-		614 0x I	1b.	I.	34	0	34	5_		
21 1	16	16	2)	15 23	17	22	1)	21	1)	20	1	21	1	19	****		Ofal	oove	K		0	<u>10</u>	1 6		
22 6 42	17	18	2)	14 25	2)	15	2)	13	2)	13	2	14	2	13	-		572 do.	1b			0	30	0		~ · · · · · ·
23 6 41	. 15	16	2)	16	2)	14		12	2)		2		2		<u>.</u>	-	713 do.	16		30	0	30	6		
24 6 39	12	14	2)	14		15		13	2)	12	2	12	2)				548 do.	1b		25	0	25	6		
25 6 44	13	13	2)		2)	14	2)	13	2)	12	2)	11	2)	12			631 do.	1b.	K			35	6		
26 6 43	19	19	2)	12	2)	13	2)	14	2)	15	2)	12	2)	13			651		I	30	0	30	6		
£7 6 42	18	17	2)	21 9	2)	13	့ 2)	13	2)	13	2	17	2	14		1	0arc 538	lb.	I	32	0	3 2	6		
28 6 41			2	20 13	2	21 12	2)	20 13	1)	19 13	2	19 15	2	$\frac{19}{14}$			do 686	1b.	I K	30	0	30	В		
29 6 42	,		2)	21 14	$\binom{1}{2}$	20 14	2)	20 13	$\binom{1}{2}$	20 11	12)	21	2)	19		- 1	do 949 do	1b.	I,	40	0	40	6		
3 0 6 38	11	12	2)	21 16	2)	15	2)	16	2)	15	12	14	2	19 15 19	<u></u>		548 do	1b.		35	C	35	6		
31 6 44			า ì	22	7 1	27	7)	20	1 1	16	2	14	12	11		_	438	_	I					204	2
												טוט	usec	u uy	uie C	Νווע	ersity	oi Pr	ewi	ıa, Ll	ыагу	ser\	vices,	∠Uʻl	J

Remarks Index to the Foregoing Table (6.)

1. Experiment No. 1:- A side of beef weighing 248 lbs. was frozen for 24 hours. In that time it was found that the beef was by no means frozen through, although freezing had penetrated a considerable distance into the deeper tissues. Twenty measles were removed from the deeper shoulder and thigh muscles. Our subject, W. H. G., swallowed four in two tubes according to our modification of Keller's method. All four scolices evaginated.

Contemporary tests were done as follows, in an incubator with governed temperature of 38°C:-

In 5% sodium taurocholate solution, 9 out of 12 scolices evaginated.

In 30% ox bile-saline solution, 1 out of 4 scolices evaginated.

Experiment No. 4:- A relatively heavy (361 lbs.) side of beef was frozen for 24 hours. Here again, freezing was not quite complete.

Our subject swallowed 10 measles in 4 tubes. All scolices evaginated.

Experiment No. 7:- A side of beef weighing 245 lbs. was frozen for 24 hours. The same remarks apply, as above. Subject swallowed 8 cysticerci in 4 tubes. All scolices evaginated.

Experiment No. 21:- An ox head containing some 20 viable measles was frozen for 24 hours. Ten of these measles were selected, since they were "covered" up by the flaps of the masseteric incisions. Our subject swallowed these 10 and all were found to be dead.

Summary:- Twenty four hours' freezing of beef carcasses split into halves (sides of beef) is not sufficient to kill <u>C. bovis</u>, since, in that time the freezing process has not had sufficient time to permeate into the deeper musculature. Shallowly situated measles such as those in the masseters are killed with ease after 24 hours' freezing.

For Illustrations see Figures 7 & 10



Fig. 7.

Fresh C. bovis scolex evaginated in two hours in 5% Sodium taurocholate solution. Magn. 7 X.



Fig. 8

Fresh C. bovis scolex eavaginated in 2 hours in 30 % ox bile-physiological saline solution. Magn. 7 X



Fig. 9.

C. bovis scolices evaginated by Keller's method after 24 hours' freezing. Magn 7 X.



Fig. 10 a.

C. bovis scolex evaginated by Keller's method after 24 hours' freezing. Magn 7 X.

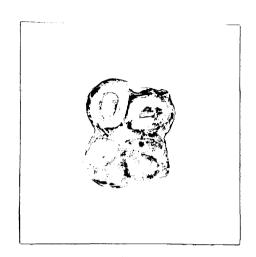


Fig. 10 b.

C. bovis head, from scolex illustrated in Fig. 10 a. Note four suckers plainly visible. Magn. 40 X.

2.

Experiment No. 2.:- Side of beef weighing 262 lbs. was frozen for 48 hours. At the end of that time the temperature of the deeper tissues of the buttock had reached 27°F. (approximate-ly-3°C.), and the room temperatures had fallen from 17°F. to 13°F. (approximately -10.5°C.) Freezing had, by that time, permeated almost completely into the deeper tissues.

A remarkable phenomenon occurred in this test, in so far as that neither of two <u>cysticerci</u> swallowed by our subject evaginated their scolices, whereas in 5% sodium taurocholate solution 4 out of 10 scolices evaginated in 2 hours, and in 30% ox bile-physiological saline solution 3 out of 10 scolices evaginated in 2 to 4 hours.

Experiment No. 6 :- A side of beef weighing 254 lbs. was frozen for 48 hours. Same remarks regarding freezing apply in this case as above. One out of 12 scolices evaginated by Kellers method.

Experiment No.3 :- An ox tongue was frozen for 48 hours. Freezing was definitely complete. No scolices from 8 measles evaginated by Keller's method.

Summary: After 48 hours' freezing in sides of beef weighing approximately 260 lbs., the freezing process has permeated considerably into the deeper musculature. The freezing action has, however, not yet had sufficient time to destroy the viability of <u>Bobovis</u>. In small pieces or meat, for example an ox tongue, the freezing process is definitely complete, and few measles will survive the low temperature reached in the interior of such meat.

Illustrations see Figures 11 and 12.





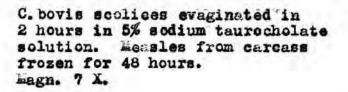




Fig. 12.

C. bovis scolices evaginated in 2 hours in 30% ox bile- physiclogical saline solution. Measles from carcass frozen for 48 hours. Magn. 7 X.

Experiment No. 5:- A fairly heavy (350 lbs.) side pf beef 3. from the same carcass as that of Experiment No. 4, was frozen for three days. After three days' freezing the inner temperature of the buttock, read on a "spear thermometer", registered 20°F. (approximately -7°C.) By that time freezing was complete. After 24 hours' thawing of the meat, WH.G. swallowed 7 6. bovis in three tubes. All seven scolices evaginated. Contemporary artificial evagination tests were tried. Five Chovis were treated in each of a 5% sodium taurocholate solution and a 30% ox bile-saline solution. Negative results were obtained. Experiment No. 8 :- A side of beef weighing 250 lbs., from the same carcass as that described in Experiment No. 7, was frozen for 3 days. The inner temperature of the meat fell from 25 F. after 24 hours to 23 F. after three days. Freezing room temperature varied between 16°F. and 11°F. Freezing was complete. Out of 12 measles swallowed by W.H.G. in four tubes, no less than 9 evaginated their scolices beautifully. Contemporary artificial tests with 10 measles in each sodium taurocholate solution and 30% ox bile-saline solution, gave negative results. Experiment No. 9 :- A side of beef weighing 248 lbs. was completely frozen through in three days. Inner temperature of the meat fell from 28°F. after 24 hours to 23°F. after 3 days. Room temperature fell from 18°F. after 24 hours to 11°F. after 3 days. Out of 10 measles swallowed by W. H. G., two evaginated

Summary: Three days' freezing of sides of beef is not sufficient to kill C.bovis. A temperature of 23°F. is easily maintained

their scolices.

in the meat after 24 hours, provided the room temperature is kept below -10° C.

Illustrations see Figures 13 and 14.

Experiment No. 10:-A side of beef weighing 300 lbs. was frozen through for 4 days. The internal temperature of the meat registered 28°F.after the first 24 hours' freezing, and fell rapidly between the third and fourth days from 23°F.to 14°F., which temperature was the same as that of the freezing chamber. On the third day the temperature of the freezing chamber necessarily rose to 19°F., as the result of the opening of the chamber for about an hour, in order to take out, and place in a few lightly infested measly carcasses for Regulation freezing treatment. After 24 hours' thawing of the meat, W.H.G. swallowed three tubes containing 10 measles. Of this number four scolices evaginated. Contemporary tests with sodium taurocholate solution and ox bile-saline solution gave negative results.

Experiment No. 12:- A hind-quarter of beef weighing 160 lbs. was frozen for 4 days. The initial temperature of the freezing chember was 21°F., but after the quarter had been in the chamber 48 hours, the temperature had fallen to 16°F., and this was succeeded on the third and fourth days by 11°F. and 15°F. respectively. The internal temperature of the meat after the 2nd, 3rd and 4th days was 23°F., 19°F., and 20°F., respectively. Freezing of the quarter was complete. After 24 hours' thawing of the meat, W.HG. swallowed 10 cysts, of which number 4 evaginated scolices. Experiment No. 18:- A side of beef weighing 281 lbs. was frozen for 4 days. Freezing was complete, and on no occasion during that

time did the freezing room temperature exceed a maximum of 16°F. (approximately =9°C.) The internal temperature of the meat fell from 23°F. (approximately -6.5°C.) after the first 48 hours' freezing to 18°F. (approximately -8°C.) after the 4th days' freezing. After 24 hours' thawing of the meat W.H.G. swallowed seven measles in 3 tubes. Of this number two evaginated their scolices. Contemporary tests with sodium taurocholate solution and bile-saline solution gave negative results.

Summary: Out of a total of 27 measles from meat frozen for four days, swallowed by W.HG., ten were still capable of evaginating their scolices under conditions tantamount to natural infection. Freezing in all cases was complete, and a uniformly low temperature was maintained. Cysticercus bovis is not destroyed by thorough freezing in all cases within four days.

Illustrations see Figure 15.

Experiment No. 11:- A side of prime "young" beef weighing 288 lbs., and containing a large quantity of fat, uniformly distributed, was frozen for 5 days. This side of beef was from the same carcass as that of Experiment No. 10. That freezing was complete can be gleaned from the following temperature records, which are specially repeated in this case:-

Te	nitial mp. of reezer.	Temp. after 5 hours	Temp. after 24 hours	Temp. after 48 hours	Temp. after 3 days	Temp. after <u>4 days</u> .	Temp. after <u>5 days.</u>	
) Of Meat) Of Chamber	19 ⁰ F.	14°F.	28° _F	22 of. 12 ^o f.	23°F. 19°F.	14°F. 14°F.	12°F.	

Only on one occasion, as was noted under the description of $E_{\rm x}$ periment No. 10, did the freezing room temperature exceed -10° C, for the reasons stated. It took forty-eight hours before

sufficient thawing had occurred to permit of the side of beef being dissected in order to collect deep-seated cysticerci.

Thirteen measles were given to W.H.G. to swallow in 4 tubes. Of this number 1 evaginated its scolex. I sent this evaginated scolex to Dr. H.O.Monnig, Onderstepcort, with a request that he pull examine and describe its condition, and, if necessary prepare it for photographing purposes. Dr. Monnig kindly favoured me with the following report).

"Scolex of C. bovis evaginated after 5 days' freezing. The scolex is provided with two apparently normal suckers situated on two adjoining quarters. The other two quarters are slightly thickened, as if rudimentary suckers were present, but no such structure is visible. Between the latter two quarters there is a large invagination of which the opening is as wide as a normal The lumen of the invagination is then constricted, but widens out again to form a fairly large space in the centre of the scolex, from which two arms are given off in the directions of the two normal suckers and almost reaching these. The orifice of the invagination is lined with cuticle bearing a number of evenlyspaced striations directed from without inwards. The walls of the invagination within the scolex are thrown into a number of large folds. It is problematic whether the scolex could have attached itself normally to the intestinal wall and, if it could have done so, it is probable that the tapeworm would have had some difficulty in maintaining its position after it had grown to some length." Illustration of this scolex, see Figure 16.

Experiment No. 15:- Side of beef weighing 250 lbs. was frozen for 5 days. Freezing was again complete and uniformly low temperatures were maintained. W.H.G. swallowed 11 measles, of which number none evaginated scolices.

Experiment No. 19:- Side of beef weighing 289 lbs. was frozen for 5 days. This side was from the same carcass as that of Experiment No. 18. Out of 11 measles swallowed, no scolices evaginated.

Experiment No. 20:- Carcass of beef weighing 614 lbs. was completely frozen through for 5 days. Note the low temperatures maintained of (1) the meat and (2) the freezing chamber. After 48 hours thawing of the meat W.H.G. and two others swallowed 34 measles. Of this number no scolices evaginated. (One bag containing 3 measles was "lost".)

Summary: Out of a total of 69 measles swallowed by my assistants, one evaginated its scolex, after five days' freezing. Dr Monnig has aptly described the deformities of the head of this evaginated scolex, but, nevertheless, if we must accept the theory of complete evagination of the scolex as the best criterion of viability of measles, then we must bear in mind that occasionally viable cysticerci may survive in beef frozen for five days.

6. Experiments 13 and 16:- Sides of beef weighing 289 lbs. and 316 lbs. respectively, were completely frozen through for six days. Out of a total of 27 measles swallowed by my assistant, WH.G., no scolices evaginated.

Experiments 22 to 31: Having established the possibility that

C. bovis might occasionally survive five days' continuous freezing,

I decided to confine my tests after the twentieth experiment to measles from carcasses frozen for six days. Consequently, the last

ten carcasses we obtained during the months of December, 1936 and January and February, 1937, were subjected to six days? continuous freezing. As many cysticerci as possible were collected from both sides of those respective carcasses, after the latter had been sufficiently thawed. The cysticerci were enclosed in tubes and silk bags and were swallowed by W.H.G. and two assistants. W.H.G, as usual recovered all his tubes, but a few of the silk bags were "lost" by his two confreres. Nevertheless, out of a total of 317 measles collected from 10 carcasses, no scolices evaginated.

7. Experiments 14 and 17:- Two fairly large sides of beef, weighing 290 lbs. and 312 lbs., respectively, were completely frozen for 7 days. Out of a total of 21 measles swallowed by W.H.G., no scolices evaginated.

The following short table summarizes the results of our freezing tests with <u>C. bovis</u>. (Sides, Quarters and Carcasses of Beef, only):-

Days Froz e n	Total Measles Swallowed	T _o tal Scolices Evaginated	Digested or "Lost"	Percentage Viable.
1	22	22	ô	100
2	14	1	13	7.1
3	29	18	11	62.07
4	27	10	17	37.04
5	69	1	68	1.45
6	344	0	344	•
7	21	0	21	-



Fig. 13.

C. bovis scolices evaginated by Keller's method after 3 days' freezing. Magn 7 X



Fig. 15.
C. bovis scolices, evaginated by
Keller's method after 4 days'
freezing. Magn. 7 X.



Fig. 14.

C. bovis scolices evaginated by Keller's method after 3 days' freezing. Magn. 7 X.

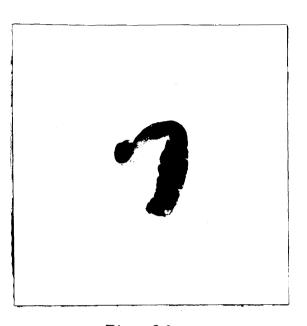


Fig. 16 a.

C. bovis scolex evaginated by Keller's method, after 5 days' continuous freezing. Magn. 7 X.



Fig. 16 b.

Microscopic view of same scolex. See description of Experiment No. 211.

Magnification 40 X.

Conclusions.

From the results we obtained with the available material at Bloemfontein Abattoir, it is reasonable to conclude that <u>C.bovis</u>, frozen in whole sides of beef can withstand a considerable amount of freezing. That the beef <u>cysticerci</u> can remain Viable under those circumstances, after 24 hours' to 4 days' continuous freezing, is without much doubt, if the fullest recognition of our criterion of simulating natural infection is to be accepted as the most conclusive evidence thus far provided by modern science.

Our table, giving the most meticulous daily temperature recordings, shows that the most thorough through freezing of the tested carcass was applied.

After five days' continuous freezing, on one occasion, my subject recovered an evaginated scolex. This scolex, as was described by Dr. Monnig, was to a great extent deformed in the head process, and only two normal suckers could be noticed. Since malformations are not altogether unusual in <u>cysticerci</u>, it may be considered possible that this particular <u>cysticercus</u> may have evaginated a deformed scolex under normal and fresh conditions. It may also not be unreasonable to presume, that since one deformed <u>cysticercus</u> out of a not excessive number tested (69), was capable of evaginating its scolex, a few quite normal <u>cysticerci</u> might be able to do so, if swallowed by humans with the ingestion of measly beef frozen for five days. We may, therefore, reasonably presume that a five days' freezing treatment is risky, if we are to satisfy ourselves of the

shortest period of freezing necessary for the destruction of Cysticerci bovis.

Superficial <u>Cysticerci</u> <u>bovis</u>, e.g. those situated in the tongue, or in the masseters of the face, are destroyed within 48 hours, according to our tests. Provided a temperature of -10°C. is maintained for that period, through freezing of a relatively thin muscular organ, like the tongue, or of a thin flap-like muscle, like the masseter, will occur with certainty.

Judging from our tests, it would appear that no <u>Cysticerci</u> bovis, situated in the deep musculature of the shoulders or of the thighs can survive a period in excess of five days' freezing. After 6 days' freezing, 344 deep-seated measles (shallow measles were ignored and not used) failed to evaginate their scolices, when swallowed according to our modifications of Keller's and Iwanizky's methods. Similarly, we had negative results with 21 measles tested on two occasions after 7 days' freezing. If our results, offered not in a dogmatic manner, were to be accepted, it can reasonably be presumed that no <u>cysticerci</u> will remain viable after six days' continuous freezing, but there is a definite probability that a few individual deep-seated <u>cysticerci</u> can survive a five days' freezing.

The South African Regulations provide for a period of 14 days' continuous freezing at -10°C., and there is no doubt at all, that this period is perfectly safe from the public health point of view. If any good purpose were to be served thereby, e.g. economy to the butchers, I feel that the period of freezing can with a margin of safety, be reduced to 10 days' freezing. The only

objection I have to a ten days! freezing process is that, unless the abattoir is a large concern and several freezing chambers may be available, it will be difficult to organise control and maintenance of low temperatures. This can be elucidated by the following example. At Bloemfontein we only have one freezing chamber available at the present time. In order to maintain the low temperature required, we have set aside one morning a week (every Thursday), when we take out of, or place into, the freezing chamber the week's number of lightly infested carcasses, which, meanwhile, have been collected and kept in a part of the chill-rooms set aside for that purpose. This operation takes about an hour every week, and after that the freezing chamber is kept locked until the next week, except for a few minutes on certain occasions wing the past year, when we were conducting our viability experiments. By setting aside a certain day once a week for working in the freezing chamber, a regular control can be exercised, whereas with ten days' freezing, irregular days of taking out or putting in carcasses will follow.

In agreement with Keller, one must stress the fact that low temperatures must be maintained, otherwise, as Keller found, all cysticerci may not be killed even after about 23 days' freezing at such high temperatures of e.g. -1°C. to -1.5°C. (± 30°F°)

COMPARATIVE TABLE SHOWING THE RESULTS AND RECOMMENDATIONS OF VARIOUS WORKERS Temperatures and Periods of Freezing Lethal to Cysticerci.

	CYSTICERCUS	CELLUL		CYSTICE	RCUS BO	OVIS.
Investigator	Temperature Required	Time	Part of Carcass	Temperature Required	Time	Part of Carcass
eissmann (1897	7) -80C. to -10°C.	4 days	depths	-8°C. to	3 days	depths.
Boccalari (1903)	-4°C. to	4 days	-	-4°C. to	4 days.	•
lans om (1914)	10	•••	_	-9°C. to	6 days	quarters
lillisch(1923)	-8°C, to -12°C.	3½ days	½ pigs	-	•	
agner (1922)			_		8 days	carcasses
chmey & ugge (1930)	-8°C. to	4 days	carcasses	-8°G. to	4 days	carcasses
allert(1931)		•		do.	9-10 days	hind quarters
larenburg (1932)	-			do.	65 hours	6 cm. pieces
eldforth (1934)				-2°c.	2 days	½ lb. weight
(X:1) cheerer (1935)			-	-2°c.	6 days	inner- most parts.
(X 2) unker (1935)	_	The state of the s	•	-3°C.	6-7 days	do.
loemfontein battoir (1936-37)	-10°C	5 days	carcasses	-10°C.	6 days	carcasses

- (X 1) Scheerer explained that a temperature of -2°C. must be maintained for 6 days in the innermost parts of the carcass.
- (X 2) Zunker maintained that a temperature of -3°C. in the innermost parts of carcass was lethal to <u>C. bovid</u> and it generally took 6 to 7 days to reach -3°C. in those parts.