

Observations were made daily as long as the animal survived.

The effects on the motility of the intestine of insulin alone, the combination of insulin and glucose, glucose alone, glucose preceded by insulin, and insulin preceded by glucose were observed. Blood sugar determinations were also made. The results obtained were consistent as regards the action of glucose and insulin on the gut. There was little difference between the reactions of the normal and obstructed gut to the insulin and glucose except that possibly insulin alone exerted a slightly more marked effect on normal gut and that which had been obstructed for 24 hours than that which had been obstructed for longer periods of time. The total results obtained are given in Table I.

TABLE I.

Solution	No. of observations	Increase in intestinal activity	No change	Decrease
		%	%	%
Insulin alone	17	58.8	35.2	5.9
Insulin + glucose	10	40.0	50.0	10.0
Glucose alone	18	0.0	11.1	88.8
Glucose preceded by insulin	37	75.6	13.5	10.8
Insulin preceded by glucose	7	71.4	28.5	0.0

Conclusion. From these results, it is suggested that glucose alone should not be used postoperatively and certainly not in the presence of intestinal obstruction, because glucose alone in the majority of instances exerted an inhibiting effect on intestinal activity. Insulin, or the combination of insulin and glucose produced an increase in intestinal activity. The best results were obtained when insulin preceded the administration of glucose by about one-half hour's time. There was no correlation between the intestinal activity and the blood sugar findings.

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Comparative Study of the Rabbit Leucocytic Count Following Injection of Various Antigenic Substances.*

WILLIAM H. HARRIS AND HERBERT J. SCHATTENBERG.

From the Department of Pathology, College of Medicine, Tulane University.

The reactions described here bear a relationship to the subject of nonspecific protein therapy although there is no intention at this time

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to consider this phase of the subject. In connection with the study of nonspecific protein reactions, many observations have been reported upon the leucocytic response especially in the human being, and particularly following the intravenous injection of *B. typhosus*. A full review is given by Petersen.¹ Ling² reported various blood reactions both in the human and in the rabbit, employing several substances for injection, including milk, peptone, horse serum and *B. typhosus*. Most of the injections were administered intravenously. He obtained only a very slight response in his leucocytic reactions.

We wish to report the comparative leucocytic reactions produced in the rabbit by the subcutaneous, intraperitoneal and intravenous routes of injection for 8 different substances.

Twenty-four full grown rabbits, chiefly males, were used and the following antigens employed: Cysteine, peptone, globulin, market milk, *Staphylococcus aureus*, *Streptococcus hemolyticus*, *B. typhosus* and the supernatant portion of a sero-purulent exudate obtained from a streptococcal arthritis. The latter was centrifugalized and the supernatant portion used as such and also after filtration through a Seitz filter. The unfiltered portion was found to contain a few scattered chains of living streptococci.

The bacterial suspensions employed contained approximately one billion bacteria per cc. and were killed with heat at 56°C. for one hour. Heat was used in order to avoid a confusion of the reaction by the addition of a chemical germicide. The other materials were used in a 2% strength with the exception of the milk and arthritic exudate which were employed undiluted. The animals of one series received 2 cc. subcutaneously, in a second series 2 cc. were given intraperitoneally and in the third series 0.5 cc. was administered intravenously excepting for *B. typhosus*, wherein 0.1 cc. was given. Larger amounts of *B. typhosus* given intravenously produced death.

Before inoculation the leucocytic count was made on all rabbits at varying intervals and of 200 estimations made from the marginal ear vein, an average of 8,000 cells per c.mm. was found. This amount is similar to the observations of others as cited in the following article. Our average differential count of the leucocytes was: Neutrophiles, 30.8%; lymphocytes, 64.4%; eosinophiles, 2.2%; basophiles, 2.4%; large mononuclears, 0.2%. We believe that in the estimation especially of total values the phenomenon of local

¹ Petersen, W. F., in Jordan and Falk, *The Newer Knowledge of Bacteriology and Immunology*. University of Chicago Press, March, 1929, 1086.

² Ling, C. Y., *Arch. Int. Med.*, 1925, **35**, 598, 740, 752.

leucocytosis described in the following article must be considered for blood obtained from the ear veins of the rabbit. We noted in some instances that when the ear was pricked repeatedly and the blood obtained with difficulty because of a poor vein, blood collected 6 hours later showed apparently a marked constitutional rise but when checked on the opposite ear, a normal result was obtained.

The results of the inoculations given subcutaneously, intravenously and intraperitoneally are recorded in Table I. From this can be seen the effects produced upon the total and neutrophilic leucocytic count provoked by the antigenic substances.

The counts were made before the injections and subsequent estimations after 6 and 24 hours. In nearly all instances the rise subsided within 24 hours. When it persisted upon the following day,

TABLE I.

Inoculum	Route	Total Count Neutrophiles $\times 100$			
		Before	After	Before	After
Cysteine	Subcutaneously	60	112	%	%
Peptone	"	54	88		*
Globulin	"	66	72		*
Milk	"	90	86		*
Arthritic Exudate					
Filtered	"	66	88		*
Arthritic Exudate					
Unfiltered	"	84	112	30	44
<i>Staph. aureus</i>	"	98	116		*
<i>Streptococcus hem.</i>	"	84	74		*
<i>B. typhosus</i>	"	86	70		*
Cysteine	Intraperitoneally	80	76		*
Peptone	"	78	128	14	21
Globulin	"	58	186	26	42
Milk	"	90	176	24	20
Arthritic Exudate					
Filtered	"	84	98	28	32
Arthritic Exudate					
Unfiltered	"	76	176	36	48
<i>Staph. aureus</i>	"	54	336	18	82
<i>Streptococcus hem.</i>	"	64	126	10	44
<i>B. typhosus</i>	"	106	304	24	70
Cysteine	Intravenously	66	116		*
Peptone	"	70	82	46	68
Globulin	"	62	188	28	52
Milk	"	86	208	40	74
Arthritic Exudate					
Filtered	"	64	80		*
Arthritic Exudate					
Unfiltered	"	64	164	21	48
<i>Staph. aureus</i>	"	126	218	16	32
<i>Streptococcus hem.</i>	"	48	66		*
<i>B. typhosus</i>	"	80	240	50	84

* Indicates no increase in neutrophiles.

the counts were continued until normal was reached. The recorded results following the injections show the peak reached by the leucocytes and represent the 6-hour period. The counts showing increase were controlled by estimations on the opposite ear.

In the rabbits injected intraperitoneally, the elevation produced by milk and peptone subsided in 48 hours, that produced by unfiltered arthritic exudate in 72 hours while the leucocytosis produced by *B. typhosus* continued for 4 days.

In the rabbits injected intravenously, the increased count subsided in 24 hours for all animals except for unfiltered arthritic exudate, *Staphylococcus aureus*, *Streptococcus hemolyticus* and *B. typhosus*. *B. typhosus* produced by this route a leucocytosis lasting over 5 days whereas for the other 3 agents it subsided in 48 hours.

The rabbits injected intravenously and intraperitoneally with *B. typhosus* appear somewhat sick for 24 to 48 hours. The leucocytic reactions in such animals are high and more lasting than for the other substances.

In the rabbits injected subcutaneously only comparatively slight or no reaction was obtained with the dosage employed.

In regard to the differential counts, as shown in Table I, the neutrophils are increased 7 to 64% in the animals showing the more marked leucocytosis. As might be expected, the greatest neutrophilic increase occurred for *Staphylococcus aureus*, although *B. typhosus* also provoked considerable increase.

TABLE II.
Increment of Leucocytes $\times 100$.

	Method of Injection		
	Subcutaneously	Intraperitoneally	Intravenously
Cysteine	52	—	50
Peptone	34	50	—
Globulin	—	128	126
Milk	—	86	122
Arthritic Exudate, Filtered	22	—	—
Arthritic Exudate, Unfiltered	28	100	100
<i>Staphylococcus aureus</i>	—	182	92
<i>Streptococcus hemolyticus</i>	—	62	—
<i>B. typhosus</i>	—	198	160

— = Increment less than 2,000.

Table II presents the total increment of the leucocytes wherein the rise was over 2,000 cells per c.mm. and also shows the comparative results obtained by each route of injection, *i. e.*, subcutaneous, intravenous and intraperitoneal.

The rectal temperatures of the majority of the rabbits with marked leucocytosis showed a rise of from 1 to 4°.

The tests described demonstrate that of the substances used for injection, consistent leucocytic increase was obtained by intravenous and intraperitoneal administrations of globulin, milk, unfiltered arthritic exudate, *Staphylococcus aureus* and *B. typhosus*. The intraperitoneal injections yield as a group the highest estimations.

These experiments represent preliminary tests to determine certain reactions relative to the resistance of infection, the study of which is now in progress.

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Production of Local Leucocytosis in the Rabbit by Mild Provocative Measures.*

HERBERT J. SCHATTENBERG AND WILLIAM H. HARRIS.

From the Department of Pathology, College of Medicine, Tulane University.

Many reports describing the normal leucocytic findings in the rabbit have been published. (Pearce and Casey,¹ Bushnell and Bangs,² Kleineberger and Carl.³) The estimations of these workers are tabulated below:

TABLE I.

	Total Leucocyte Count	Neutro- philes	Lympho- cytes	Large Mononu- clear	Eosino- philes	Baso- philes
		%	%	%	%	%
Bushnell and Bangs....	10,657	39.1	53.9	0.43	1.1	3.4
Pearce and Casey	9,562	45.4	31.8	10.5	2.2	9.9
Kleineberger and Carl	8,150	50.5	35.0	10.5	1.0	2.5

The findings of Scott and Simon and of Cunningham, Sabin, Sugiyama and Kindwall are cited by Pearce and Casey.

In our observations upon the normal count of rabbits, the blood from the marginal ear vein of 27 normal full grown rabbits, mostly males, was employed. Two hundred counts were made from these animals, and the following estimations obtained:

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¹ Pearce, Louise, and Casey, A. E., *J. Exp. Med.*, 1930, **51**, 83.

² Bushnell and Bangs, *J. Inf. Dis.*, 1926, **39**, 291.

³ Kleineberger and Carl, cited by Kolmer and Boerner, *Approved Laboratory Technic*, 1931, **65**. D. Appleton & Co., New York and London.