intensity of stimulus adequate for this purpose in cats with normal body temperature (11). Also, while electrical stimulation of the motor cortex elicits peripheral movement of the golden hamster during arousal from hibernation, the threshold declines as the animal warms(12).

Summary. The increased brain excitability observed in cold-exposed, intact rats is not mediated through pituitary hyperfunction, because hypophysectomy increases this effect rather than hindering it. In hypophysectomized, cold-exposed rats, the lowering effects on electroshock seizure threshold of absence of ACTH and mineralocorticoids predominate over the opposite effects of hypothermia and absence of thyroid and glucocorticoid hormones.

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Non-Competitive Non-Inhibitory Interaction Between Hippurate and Creatinine in the Kidney.* (28992)

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Under appropriate conditions, renal excretion of creatinine chromogen in several animal species exceeds its glomerular filtration(1). This phenomenon recently has been confirmed in the dog and has been attributed to renal tubular activity(2,3). Inhibition of creatinine excretion by 4-aminohippurate also has been demonstrated and has been described as a competitive phenomenon(3) on the assumption that creatinine and 4-aminohippurate share a common transport mechanism. The following experiments with renal cortical slices do not confirm the contention that hippurate and creatinine share a single transport mechanism.

Methods. Male albino rabbits weighing from 1-2 kg were decapitated and exsanguinated, and their kidneys were removed. Slices of rabbit kidney cortex were prepared with a Stadie-Riggs microtome and were incubated in a phosphate-buffered saline medium (pH 7.4) at 27°C for one hour in an atmosphere of oxygen(4). At the end of incubation the concentration of creatinine chromogen(5) or of 4-aminohippurate(6) both in slices and in media was determined for calculation of slice to medium concentration ratios.

Results. Concentration ratios for creatinine chromogen. Data are summarized in Table I. Each figure is the arithmetic mean of a total of 9 observations in 3 rabbits. Since acetate and probenecid had no significant influence on the concentration ratios for creatinine chromogen, there is no evidence that creatinine is actively accumulated by renal cortical slices. It is, therefore, unlikely that creatinine and 4-aminohippurate share a single transport mechanism.

Interaction of creatinine chromogen and 4-aminohippurate. Data are summarized in Table I. Since creatinine did not influence

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TABLE I. Slice to Medium Concentration Ratios.

Substance (conc)	Control	Acetate (.01 M)	Probenecid (.05 mM)	Anoxia
Creatinine (.1 mM)	.93	.94	.87	.92
" (.2 mM)	1.1	1.1	.98	.89

	Creatinine		
	Control	.3 mM	3.0 mM
4-aminohippurate (.05 mM)	5.9	5.8	—
	9.9	—	10.1

the accumulation of 4-aminohippurate, neither competitive nor non-competitive inhibition could be demonstrated.

Generation of creatinine chromogen by renal tissue. Fresh slices of rabbit kidney cortex contained pre-formed creatinine chromogen in a concentration of 17 μg per gram of tissue. At the end of the incubation period, total recovery of creatinine chromogen from slices and media was equivalent to a concentration of 35 μg per gram of tissue. The increase of creatinine chromogen during incubation was unaffected by probenecid (0.05 mM) or by anoxia.

Discussion. Competition between two substrates during renal transport has been a frequent claim although data which support such claims rarely satisfy kinetic criteria for competition. In the present instance, 4-aminohippurate and creatinine are said to compete with each other because the clearance of creatinine is depressed by concurrent administration of hippurate(3). Substantiation of such a claim, requires as additional minimum demonstrations first, that the two compounds share a single transport mechanism and second, that they are mutually inhibitory during transport. However, creatinine is not concentrated by renal cortical slices under conditions which are sufficient for the concentration of 4-aminohippurate. In addition, accumulation of creatinine by kidney slices does not respond in the same way as does accumulation of 4-aminohippurate to the influences of acetate, probenecid or anoxia. These observations permit the conclusion that creatinine and 4-aminohippurate do not share a single transport system. Furthermore, although 4-aminohippurate inhibits excretion of creatinine(3), creatinine has no effect on the transport of 4-aminohippurate; consequently, the two compounds cannot be described as being mutually inhibitory. It is probable, therefore, that the interaction between 4-aminohippurate and creatinine which has been observed in dogs(3) is unrelated to any involvement of the hippurate transport process. As a result, apparent tubular transport of creatinine as well as its response to the administration of 4-aminohippurate must be accounted for in other terms.

Perhaps creatinine clearances which exceed inulin clearances(2,3) are representative of tubular secretion in which creatinine is synthesized in the kidney cell before being transported into the tubular urine. This contrasts with the commonly expressed view that in appropriate species creatinine is transported from the blood to the urine by tubular excretion(2,3). The demonstration that creatinine chromogen is produced by rabbit kidney slices during both aerobic and anaerobic conditions supports the former view. The source of this chromogen remains obscure. Kidney tissue has been shown to synthesize glycocyamine, a creatinine precursor(7); however, conversion of glycocyamine to creatine, an intermediate in the pathway to creatinine, appears not to take place in the kidney. Production of creatinine chromogen in the present experiments possibly might be attributable to spontaneous alteration of preformed creatine. Such a reaction in isolated rabbit tissue under the conditions used is neither very large quantitatively nor is it sensitive to probenecid and anoxia. Nevertheless, it is conceivable that conditions in intact experimental animals might well be adequate for the production of significantly large quantities of creatinine chromogen. A

parallel phenomenon, the renal synthesis of urea, has been described(8). Confirmation that the kidney can also serve as a source of creatinine might reorient interpretation of available experimental observations.

Summary. In slices of rabbit kidney cortex, active transport of 4-aminohippurate was uninfluenced by creatinine even when the concentration of creatinine was 60 times greater than that of hippurate. Previously published claims that creatinine and 4-aminohippurate compete for a single transport mechanism are thereby refuted. A small but significant rate of synthesis of creatinine chromogen by renal tissue conceivably can serve as a renal source of creatinine during clearance measurements.

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Plasma Enzymes Erythropoiesis and R.E.S. Function in Mice Following Infection with an LDH Agent.* (28993)

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Riley(1) reported that a number of murine tumors were contaminated with a virus-like agent which could be propagated independently of tumor by serial passage of plasma from infected mice. The agent was detected only by a sharp rise of 5- to 10-fold in plasma lactic dehydrogenase level about 2 to 3 days following infection(2). General properties of the agent, as described by Riley, have been confirmed by a number of workers (3,4,5). An agent having certain similarities to LDH agent was reported by Old *et al*(6) in mice bearing sarcoma 180. This agent caused hyperactivity of the reticulo-endothelial system. No observations of plasma LDH were reported.

In a previous study(3) no difference in LDH enzyme content of a number of tissues of both normal and agent-infected mice was found. There was, however, a slightly but significantly lower hematocrit in infected mice which suggested red blood cells as a possible source of elevated plasma LDH.

As a possible means of identifying the target tissue for the LDH agent, therefore, it was of interest to determine levels of a number of different enzymes in plasma of mice both before and after infection with agent. Similar studies have recently been reported by Notkins and by Plagemann(9,10). Erythropoiesis in normal and infected mice was studied during an intensive bleeding schedule. R.E.S. function was examined by measuring rate of carbon clearance from plasma. Gross autopsy studies were made on fixed and stained sections of liver, kidney, brain, lung, spleen and thymus of infected mice.

Materials and methods. Source of LDH agent used in this work was CD-5 lymphoma carried in BALB/c mice.† Pattern of replication of agent, demonstration of virus-like nature and similarity to agent isolated by Riley have been reported(3).

Mice of BALB/c strain were bled from

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