attachment, or by some other, as yet unknown, mechanism. Such inactivation might also serve as a possible cause for infertility among otherwise physiologically normal couples.

The object of this study has been to determine whether or not the secretions of the uterine cervix contain hemagglutinins, as an initial step in an effort to work out the possible physiological basis for ABO selection. The impetus for the study stemmed from the feeling that the known facts regarding the consequences to the fetus of ABO isoimmunization (see 7) were not sufficient to account for the magnitude of selection effects postulated for ABO incompatible matings. The detection, some 30 years ago, of hemagglutinin in the cervical fluid was apparently limited to the analysis of a *single* sample (*8*).

All the women investigated in the course of this study were patients in the Out-Patient Gynecology Clinic of the University Hospital. Their reasons for the clinic visit were varied; only five were seen because of failure to conceive and in whom the cause of infertility was still unknown after careful study of the marital partners. The collection of cervical secretion depended only on whether or not a sample could be obtained from any particular woman. Blood for typing was obtained at the same time. All secretions were frozen soon after collection and all were tested within a period of 5 days.

The method of testing was as follows. To each tube containing the sample, which was usually quite small, was added 0.15 ml of saline. Because the quantity of secretion obtained varied from subject to subject, the dilution factor is variable. The contents of the tube were then thoroughly agitated with an applicator, the tube was centrifuged, and the clear supernatant fluid was tested with fresh A_1, B, and O red blood cells. All tests were performed in microtubes (6 by 50 mm) to which 0.01 ml of a 2 percent washed red blood cell suspension was added to 0.01 ml of the diluted secretion. The tests were allowed to stand for 1 hour at room temperature and were read macroscopically both before and after centrifugation.

The results are given in Table 1. Only distinctly positive reactions are tabulated, although the inclusion of weak, ambiguous reactions would not have substantially changed the magnitude of the difference presented in the table. Fifteen of the 35 diluted secretions (42.8 percent) of blood type O women contained antibody of the ABO system. Of 30 type A and 5 type B women, two cervical secretions (5.7 percent) had detectable antibody. Hemagglutinins were not found in the secretions of seven type AB women, which is additional evidence for the ABO group specificity of these antibodies. Only one of the women with cervical hemagglutinins had been seen because of infertility. A comparison of the findings in type O women with those in types A and B by the exact treatment of a 2×2 table yielded the two-tailed probability of 0.0053 that they were drawn from the same population. The fact that the over-all percentage of O, A, and B women exhibiting detectable hemagglutinins was only 24.3 may be due in part to the degree of dilution necessary to bring the volume up to 0.15 ml for scanty specimens, but this suggestion leaves unexplained the differences between A or B and O women. To some extent the latter differences may be due to the greater opportunity to detect agglutinins in O women, who have *two* agglutinins. Studies on undiluted secretions are in progress.

These findings would appear to support the possibility of gamete selection as a means of accounting for the distortions of the expected ratios in marriages of A fathers and O mothers, a possibility which, after the completion of this study, was discovered to have been proposed by Matsunaga in 1953 (*2*). Although several further lines of investigation readily present themselves, really critical data on this point may be difficult to obtain. The observed departures from the Mendelian ratios could be produced by a relatively slight impairment of sperm motility within women of the appropriate genotype, an impairment difficult to demonstrate with certainty. Inasmuch as the increased frequency of abortions in incompatible marriages seems securely established (*2, 9*), the present findings may indicate that the departure from Mendelian ratios in the children of A male × O female marriages has a complex background.

Note added in proof: It has recently come to our attention that the demonstration of antigenic dimorphism in the sperm of an AB male has been made by Gullbring (*10*). This finding would seem to support the possibility of preconception selection by blood group antibody in the uterine secretions.

Table 1. Number of diluted uterine cervical secretions containing hemagglutinin, tabulated by blood type of the donors.

Blood type	No. tested	No. containing only anti-A	No. containing only anti-B	No. containing anti-A and anti-B	Total no. with hemagglutinins
A	30	0	2	0	2
B	5	0	0	0	0
AB	7	0	0	0	0
O	35	4	2	9	15
Totals	77	4	4	9	17

H. Gershowitz
S. J. Behrman
J. V. Neel
Departments of Human Genetics and *Obstetrics and Gynecology, University of Michigan Medical School, Ann Arbor*

References and Notes

1. J. A. H. Waterhouse and L. Hogben, *Brit. J. Social Med.* **1**, 1 (1947).
2. E. Matsunaga, *Proc. Japan Acad.* **29**, 399 (1953).
3. J. Bennet and J. Brandt, *Ann. Eugenics* **18**, 302 (1954); J. H. Edwards, *Brit. J. Prevent. and Social Med.* **11**, 83 (1957).
4. E. Matsunaga and S. Itoh, *Ann. Human Gen.* **22**, 111 (1958).
5. This investigation was supported in part by a grant from the U.S. Atomic Energy Commission [project AT(11-1)-405].
6. K. Landsteiner and P. Levine, *J. Immunol.* **12**, 415 (1926).
7. W. W. Zuelzer and E. Kaplan, *Am. J. Diseases Children* **88**, 307, 319 (1954).
8. E. M. Schwarzmann, *Z. Geburtshilfe u. Gynäkol.* **92**, 505 (1928).
9. C. McNeil *et al.*, *Am. J. Clin. Pathol.* **28**, 469 (1957).
10. Gullbring, *Acta Med. Scand.* 159, 169 (1957).

24 April 1958

Concentration of Albumin in Renal Papilla

Recent investigations (*1*) have focused attention on the renal papilla for its role in concentrating urine. Freezing-point determinations of papillary tissue have demonstrated a tissue osmolarity up to three times that of the blood or renal cortex, because of a high concentration of sodium, chloride, and urea. Completely unsuspected, however, is the observation reported in this note of the high albumin concentration in the mammalian papilla. This may well bear an important relation to water transport in the papilla as part of the mechanism of concentrating urine.

Fourteen normally hydrated, nonanemic dogs were anesthetized with pentobarbital sodium. About 500 μc of I^{131}-labeled human serum albumin was injected systemically. The kidneys were exposed through an abdominal incision, and a loose tie was placed around the pedicle. In four dogs one kidney was ligated 3 minutes, the other kidney 60 minutes, after the injection of the radioactive material. In the remaining ten dogs both kidneys were ligated simultaneously after about 1 hour. The kidneys were removed and frozen in Dry Ice. Sections were cut on a band saw, and

pieces of the papilla were removed, placed in counting vials, and weighed. The tissue samples and 1 ml of arterial plasma obtained at the time of ligation were assayed for radioactivity in a well-type scintillation counter. The papillary concentration of labeled albumin was expressed relative to that of corresponding plasma from the following formula: Concentration of I^{131} albumin = (I^{131} counts per minute per gram of papilla)/(I^{131} counts per minute per milliliter of plasma).

The results are shown in Table 1. In about 1 hour the papillary I^{131} albumin concentration corresponded to that of 0.35 ml of plasma (± 0.09 ml S.D.). In 3 minutes the corresponding value averaged about 85 percent of the 60-minute value of the same animals. Autoradiographs taken from kidneys ligated at 3 and 60 minutes were very similar and showed a considerably higher concentration of albumin in the renal papilla than in other anatomical regions of the kidney (Fig. 1).

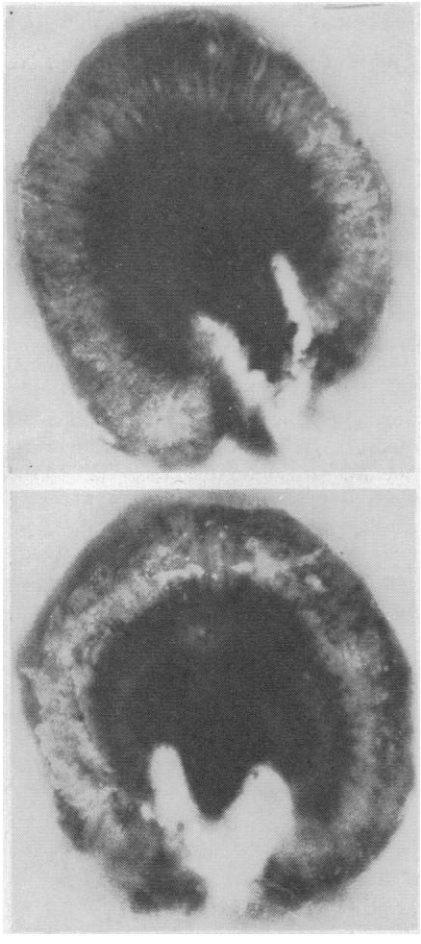

Fig. 1. (Top) Autoradiograph of a transverse slice of dog kidney obtained after the kidney was allowed to equilibrate for 3 minutes with intravenously injected I^{131} human serum albumin. Note the concentration of albumin in the papilla. (Bottom) Autoradiograph of a transverse slice of dog kidney obtained after the kidney was allowed to equilibrate for 60 minutes with intravenously injected I^{131} human serum albumin.

Urine samples collected from the bladder at the end of the experiment were hypertonic in all dogs and were found to be essentially free from radioactivity. The albumin used contained less than 2 percent of nonprotein bound radioactivity. Similar low values were found in filtered homogenates of papillary tissue. In four additional studies, labeled canine albumin was used instead of human albumin with the same result. The papillary concentrations of radioactivity were found to correspond to 0.33, 0.33, 0.52, and 0.34 ml of plasma in these four dogs. All were studied 60 mintes after injection of the radioactive material.

These data indicate a large and rapidly equilibrating pool of albumin in the renal papilla. Since complete equilibration may not have been reached in 1 hour, the values presented are minimal values for the papillary albumin concentration. Thus it is to be concluded that 1 g of the renal papilla in the dog contains at least as much albumin as is contained in about 0.35 ml of plasma. Since dog plasma contains about 3 g of albumin per 100 ml, this would correspond to an absolute albumin concentration of about 1 percent (wt./vol.). If the albumin in this pool is assumed to exist in the same concentration as that of the blood plasma, the data would indicate an albumin space of at least 35 percent of the volume of the papilla. This albumin pool is not paralleled by a large pool of erythrocytes. By direct observation the papilla is usually quite pale. Actual measurements of the red cell concentration of papillary tissue by means of Cr^{51}-labeled red cells have shown that the red cell content of the papilla is about 4 percent of its volume (*2*).

Preliminary studies in this laboratory have indicated that I^{131}-labeled human gamma globulin is incorporated more slowly than albumin into the renal papilla of dogs. This observation suggests that some portion of the papillary albumin is located extravascularly. In addition, the possibility of intravascular concentration of albumin above that of plasma must be considered. Since the papilla is exclusively perfused by postglomerular blood, a moderate concentration of intravascular papillary albumin to about one-third above that of plasma is to be expected. Histochemical observations of the renal papilla of rats have suggested that an intravascularly located plasma protein-bound esterase is concentrated several times above that of plasma (*3*). The very high values for the papillary albumin concentration found in this study are consistent with the hypothesis that the albumin in the papillary vessels is concentrated two or three times above that of plasma. If this hypothesis is correct, then water must have been removed from the plasma during its passage down into the papilla. This might be accomplished by means of the counter-current exchange of water in the vascular bundles of the renal medulla. An unusually high gradient of oncotic pressure may thus be maintained in the papilla, favoring the rapid net transport of water from the interstitial spaces into the papillary blood vessels (*4*).

Table 1. Albumin concentration in the renal papilla. In dogs 1, 2, 3, and 4 one kidney was removed 3 minutes after the injection of the radioactive albumin. The albumin concentrations in these papillae corresponded to 0.23, 0.18, 0.23 and 0.32 ml of plasma, respectively. This is about 85 percent of the corresponding 60-minute value of the same animals. For dogs 5 to 14 the values given in the table are the averages obtained from simultaneously ligated kidneys.

Dog	Time of ligation (min)	Albumin concn. relative to plasma
1	60	0.29
2	60	0.22
3	60	0.31
4	60	0.32
5	40	0.29
6	42	0.38
7	61	0.38
8	63	0.47
9	64	0.37
10	73	0.54
11	73	0.45
12	60	0.26
13	60	0.22
14	67	0.33
Av.		0.35
Standard deviation		± .09

NIELS A. LASSEN
JAMES B. LONGLEY
LAWRENCE S. LILIENFIELD
Department of Medicine, Georgetown University Medical Center, Washington, D.C.

References and Notes

1. H. Wirtz, B. Hargitay, W. Kuhn, *Helv. Physiol. et Pharmacol. Acta* 9, 196 (1951); K. J. Ulbrich and K. H. Jaramsch, *Arch. ges. Physiol. Pfluger's* 262, 537 (1956); J. D. Crawford, A. Paresi, P. Meara, M. L. Terry, *A.M.A.J. Diseases Children* 94, 512 (1957) (abstr.).
2. L. S. Lilienfield, N. A. Lassen, J. C. Rose, *Proc. South. Soc. Clin. Research* (25 Jan. 1958), p. 32 (abstr.).
3. J. B. Longley and M. S. Burstone, *J. Histochem. and Cytochem.* 6 (1958) (abstr.).
4. This work was supported in part by grant H-2987 from the National Heart Institute, U.S. Public Health Service, and was done during the tenure by Lilienfield of a research fellowship of the American Heart Association.

4 April 1958