THE ABORTIFACIENT ACTION OF THE SERUM AND URINE FROM PATIENTS WITH CANCER

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The present report describes and offers a discussion of experiments that deal with the observations of Elsasser and Wallace,² who have stated in Science: "It would seem, then, that the urine and serum of patients with malignant tumors have a selective destructive action on embryonal or newly growing tissue." These investigators briefly described an abortifacient action in pregnant rabbits produced by the intravenous injection of the serum or urine from patients with malignant tumors. This effect was not observed in rabbits by Elsasser and Wallace when they injected urine or serum from normal patients, or "anterior pituitary Antuitrin S (500 units daily) together with estrogenic hormone (20,000 units)."³

The uterine changes were described by Elsasser and Wallace as a progressive placental necrosis associated with an infiltration of inflammatory cells. The process was observed to start at the inner border of the decidual cells and it ultimately broadened to include the entire embryonal mass on the decidua. With the discharge or resorption of the fetal structures, the uterus eventually returned to a normal state. Early in pregnancy the abortifacient caused a rapid loss in the structure of the retained fetus, while later the fetal parts, without marked changes in their appearance, tended to be expelled.

In normal female rabbits the urine or serum from cancer patients was said to produce degeneration and destruction of the granulosa-cell portions of the graafian follicles, leading to sclerosis of the ovaries. When injected into normal male rabbits the alleged cancer principle produced a degeneration of the testes leading to a complete absence of the spermatogenic processes.

Elsasser and Wallace also observed that the urine from cancer patients injected into rats bearing Walker rat carcinoma 256 caused a different type of necrosis in this tumor from that which commonly occurs spontaneously. Instead of a central necrosis, which extends peripherally, the degeneration in their experiments occurred at the periphery and involved not only cancer cells but also "the epithelium of newly formed blood vessels."⁴

In résumé, the report of Elsasser and Wallace leads one to believe that patients with cancer produce and carry in their blood serum and excrete in their

³ This investigation has been aided by a grant from The Jane Coffin Childs Memorial Fund for Medical Research.


⁴ The term "anterior pituitary Antuitrin S" is interpreted by the present authors to be synonymous with anterior pituitary-like substance of the urine of pregnancy, and with the pharmaceutical product, Antuitrin S (Parke, Davis & Company, Detroit, Michigan).

⁴ Here the present authors interpret the term "epithelium" to be an error in proof-reading, endothelium having been intended by Elsasser and Wallace.

233
urine a principle that produces abortion in pregnant rabbits by way of a necrosis of the fetal parts. It also produces a degeneration of the gonads of normal animals, and a peripheral necrosis of the malignant cells and of the stromal endothelial cells in the Walker rat carcinoma 256.

These observations, could they be confirmed in whole or in part, open for study avenues whose breadth hardly needs mention to be appreciated. An attempt to confirm the above results has been made in this laboratory with the aid of Dr. Theodore H. Elsasser. The experiments now to be reported were devised in order to observe the effect on the pregnancies of rabbits of injections of serum or urine from 6 patients believed to be without cancer and 10 patients each with an advanced form of malignant disease that had been verified by an accepted diagnostic procedure. None of these patients had received any form of radiation therapy.

**Methods**

*The Test-animals:* Fifty rabbits that had previously had at least two normal pregnancies were selected under the supervision of Dr. Elsasser, and were mated on two successive days, following which they were transported in separate cages to this laboratory. On the 7th to 10th days following breeding the animals were examined by gently palpating the uterus, and those that were judged to be pregnant were given marks of identification and placed two in a cage. Their diet was a generous daily ration of oats and of fresh greens.

*The Tested Specimens of Urine and Serum:* The patients providing the specimens were admitted to the New Haven Hospital, and the fluid intake of each was limited to 1,000 c.c. per twenty-four hours. After a preliminary twelve hours of restricted fluids, the urine specimens were collected for twenty-four hours with careful supervision. With this method the output was only a few hundred cubic centimeters. When the amount was considerable, as it was on one occasion with an uncooperative patient, the collection was repeated the following day and a concentrated specimen was obtained. This portion of the experiment was carried out by Thomas Hale and B. B. Whitcomb, who submitted the specimens as unknowns for testing by K. W. Thompson and Dr. Elsasser.

Each specimen of urine to be injected was tested daily for its reaction with litmus paper, and when on two occasions it was alkaline, it was acidified slightly with glacial acetic acid. Following this, 20 c.c. were filtered once through filter paper (E. & A., white, No. 23150), in preparation for the injection. On the third day of the experiment the supply of urine was replenished with a fresh twenty-four-hour collection, or with the first specimen voided by the patient during the morning of that day.

Approximately 75 c.c. of venous blood was secured from each patient on the morning of the completion of the twenty-four-hour collection of urine. The clots were separated from the edges of the tubes, which were allowed to stand in the refrigerator until noon. Each specimen was then centrifuged, and the clear non-hemolyzed serum was bottled. The blood was handled with sterile technic so that a preservative was not required.

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5 Cancer case No. 5, urine No. 1, and cancer case No. 9.
The urine or serum was injected with a No. 25 gauge needle into a vein of a rabbit’s ear at the rate of 1–2 c.c. per minute. The amount given was: of urine, 20 c.c. once daily for four days; of serum, 6 c.c. daily for three days.

Examination of the Test-animals: The animals were sacrificed by an intravenous injection of a few cubic centimeters of chloroform on the fifth day after the first injection. Each animal was autopsied promptly before sacrificing another, and certain of the tissues were placed in fixative at once. The uteri were opened with a razor blade at a point opposite a placental site before this fixation. The fixative used was Zenker-formol; some duplicates were placed in 10 per cent neutral formalin.

Histological preparations of the tissues were made, although many of the placentae were obviously so necrotic that no detailed examination was necessary to settle this point.

Patients Providing Tested Serum and Urine

In this preliminary study the patients selected for the test had either an outspoken form of malignant disease or no detectable malignant disease. Since it is apparent that the physical condition of the patient may have had some relationship to the phenomena under consideration, there will be given in the following paragraphs a résumé of each case to indicate briefly the status of the patients whose serum and urine were tested for the abortifacient effect.

Cancer Cases

No. 1. Carcinoma of Stomach: E. C. (N.H.H. A91519), male, 67, had episodes of vomiting related to meals, loss of weight, and anorexia; duration of symptoms six months. There was a palpable mass in the epigastrium. Roentgen examination revealed carcinoma of the stomach, which at laparotomy proved to be inoperable. No biopsy was done. At time of test, three months after operation, the patient was emaciated and suffered from nausea, vomiting, and weakness.

No. 2. Adenocarcinoma of Rectum: J. R. (N.H.H. A79151), male, 72, complained of constipation, lower abdominal pain, blood in the stools, and loss of weight; duration of symptoms seven months. Roentgen examination demonstrated carcinoma of the rectosigmoid with extensive involvement of the sigmoid confirmed by sigmoidoscopy. Biopsy of the rectal mass revealed adenocarcinoma. The tumor was inoperable and a colostomy was done. At time of test, nine months after operation, the patient was emaciated and in extremely poor physical condition with progressive weakness and pain in the back and legs.

No. 3. Squamous-cell Carcinoma (Grade 2) of Esophagus; Pulmonary Tuberculosis; Syphilis: F. Z. (N.H.H. A67468), male, 54, had difficulty in swallowing, with vomiting and extreme weight loss; duration of symptoms two months; Wassermann +, Kahn +. Roentgen examination revealed obstruction and ulceration of the esophagus, as well as tuberculosis of the right lung. Biopsy demonstrated squamous-cell carcinoma, Grade 2, of the esophagus. Gastrosotmy was done. At the time of the test, three weeks later, the patient was in very poor condition, although taking nourishment via the gastrosotmy.

No. 4. Adenocarcinoma of Rectum: D. F. (N.H.H. A67935), male, 53, had a recurrence of epigastric cramps, vague abdominal pains, nausea and vomiting, with alternate diarrhea and constipation, two years and a half following a Mikulicz resection of the sigmoid for adenocarcinoma. There was a palpable polypoid recurrent tumor in the rectum shown by biopsy to be adenocarcinoma. Roentgen examination revealed no definite recurrence of
the sigmoid tumor; duodenal ulcer was also present. At time of test, one week after biopsy, the patient was in poor general condition, complaining of epigastric and lower abdominal pain.

No. 5. Carcinoma of Stomach: M. C. (N.H.H. A93824), female, 54, complained of epigastric fullness and postprandial eructations for a year. Later there occurred difficulty in swallowing, occasional vomiting, anorexia, and loss of weight. Roentgen examination revealed a typical carcinoma defect in the cardia of the stomach, with obstruction to the lumen of the esophagus. Gastrostomy was done. At time of test, two weeks later, the patient was in fair general condition, taking nourishment via the gastrostomy.

No. 6. Adenocarcinoma of Stomach: H. M. (N.H.H. 64998), male, 54, had a Finney pyloroplasty performed nine years previously for ulcer at the pylorus. Symptoms were relieved for two years, and then recurred, with signs of impending perforation of peptic ulcer. A subtotal gastrectomy was done with resection of the pylorus, including a large posterior penetrating ulcer. The pathological diagnosis was adenocarcinoma of the stomach, undoubtedly incompletely excised. At time of test, seven months after operation, the patient was gaining weight but having severe intermittent epigastric pain; a recurrence was suspected, but not definitely indicated by roentgen examination.

No. 7. Adenocarcinoma of Rectum: I. W. (N.H.H. 37107), male, 65, had obstipation for a year and a half, with occasional tenesmus and passage of blood, and acute symptoms of intestinal obstruction of two days' duration. Roentgen examination revealed carcinoma of the rectum as well as bilateral apical tuberculosis. Biopsy of the rectal mass revealed adenocarcinoma, inoperable. Colostomy was done. At time of test, one year later, the patient was gradually declining in strength, with pain and moderate loss of weight.

No. 8. Carcinoma of Stomach; Diabetes; Arteriosclerosis: E. McK. (N.H.H. 85782), male, 55, for three years had symptoms of multiple peptic ulcers, confirmed by roentgen examination; two weeks of moderately severe symptoms of intestinal obstruction, with blood in stools and epigastric pain. Roentgen rays showed prepyloric ulcer with the crater on the greater curvature, interpreted as carcinoma because of location and size. A discrete nodule in the lung was believed to be metastatic. The patient was diabetic, and a year earlier had one leg amputated because of circulatory deficiency with infection. Gastric operation was refused, and a Sippy régime instituted. At time of test, one month after the diagnosis was made, the patient was symptom-free and except for emaciation in fair condition, having a mild circulatory deficiency with healing infection of the remaining foot.

No. 9. Adenocarcinoma of Rectosigmoid: F. B. (N.H.H. A77644), male, 72, had abdominal distention, diarrhea, blood in the stools, and tenesmus; duration of symptoms one month. Roentgen examination revealed deformities diagnostic of carcinoma at the rectosigmoid junction, confirmed by biopsy; diagnosis adenocarcinoma. The tumor was inoperable and colostomy was done. At time of test, one year later, the patient was in fair condition, without symptoms, and was working.

No. 10. Adenocarcinoma (Grade III) of Colon: F. R. (N.H.H. A88361), male, 62, had epigastric pain two or three hours following meals, pain in the back, and occasional tarry stools; loss of 15 lbs. in weight; duration of symptoms four months. There was a tender movable mass in the abdomen. Roentgen examination demonstrated an infiltrative mass in the proximal transverse colon. The test was done two days preoperatively; condition excellent. Two days later tumor of the colon was verified and resected.

Non-cancer Cases

No. 1. Chronic Ulceration of Leg, following Trauma and Osteomyelitis: F. W. (N.H.H. 80042), male, 23, had a compound fracture of the leg nine years ago; intermittent ulceration and healing for the past six years with ineffectual skin grafting; chronic draining osteomyelitis following an accident three years ago; obesity, possibly due to inactivity. Operation: pedicle graft from thigh to region of ankle. At time of test, two weeks postoperative, the patient was in good general condition. Although cancer of skin is common in this type of case, it could not be demonstrated in this instance.

No. 2. Atrophic Arthritis: R. M. (N.H.H. A91522), male, 18, had pain and stiffness in both hip joints for four years; ankylosis of hips for eight months; constant traction in hospital for three months. Roentgen examination showed atrophic arthritis of hips, knees, and right sacro-iliac joint. Treatment consisted in manipulation of hip joints and physio-
TABLE I: Results of Tests of Urine and Serum from Patients With and Without Cancer:
Gross and Histologic Findings in Uterus Compared

<table>
<thead>
<tr>
<th>Cancer</th>
<th>Gross</th>
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<th>Gross</th>
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<td>N2</td>
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<td>N3</td>
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<td>N4</td>
<td>Not pregnant</td>
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<tr>
<td>N6</td>
<td>d.</td>
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+ Definite evidence of necrosis of fetal parts
- No definite evidence of necrosis of fetal parts
± Decision could not be made definitely
d. Animal died before time for autopsy
e. Early abortifacient effect
h. Extensive hemorrhage in placenta
ac. Acetic acid added to neutralize highly alkaline urine
n. Not sectioned because of post mortem changes.

therapy. At time of test, two weeks after manipulation, the patient was in fairly good general condition.

No. 3. Malum coxae senilis: A. A. (N.H.H. A58891), male, 56, had pain in lower back and in left hip, of three years' duration, with increase in severity of the pain in the hip for six weeks. Roentgen examination showed hypertrophic changes in the hip. The diagnosis was osteoarthritis. Operation consisted in acetabuloplasty with excision of anterior capsule of hip joint. At time of test, two weeks after operation, the patient's general condition was good. The hip was being subjected to traction.

No. 4. Secondary Post-traumatic Glaucoma: J. A. (N.H.H. A14573), male, 47, had secondary post-traumatic glaucoma. Irrigation of the anterior chamber of the eye was done for hyphema. At time of the test, one week after operation, the general physical examination was negative except for the eye condition.

No. 5. Cataract: W. L. (N.H.H. A73571), male, 65, had an extraction of a cataract and an iridectomy for incipient cataract. At time of test, one week after operation, the physical examination was negative except for the eye condition.

No. 6. Foreign Body in Eye: C. L. (N.H.H. A12539), male, 15, had a foreign body extracted from the eye one week before the test; general condition excellent.

Results

The following effects were observed in the 38 injected rabbits: 19 showed unmistakable gross signs of death of the fetuses, and in no instance were these found to be in error when the tissues were examined histologically; 7 showed
no signs of abortion on gross examination, but 1 of these after histologic study proved to have an early necrosis of the fetal parts; 4 were listed as doubtful on gross examination, 3 of them later proving to be necrotic, and 1 normal; 8 animals died before the completion of the test, and of the 4 whose tissues were not autolyzed, 3 showed extensive hemorrhages into the placentae, while 1 was apparently normal (Table I).

Of the tested cancer patients, 8 out of 10 had the abortifacient agent in both urine and serum; in a ninth patient it was present in the urine but not in the serum, and in the tenth case neither serum nor urine showed the effect. The abortifacient was found in neither urine nor serum of 3 of the 6 normal patients, but it was present in the serum of 2 supposedly normal patients and in both serum and urine of another (Table I).

The Observed Pathological Changes: The External Genitalia: An inspection of the external genitalia of rabbits before autopsy usually revealed some evidence of the action of the abortifacient, namely a brownish mucoid discharge and a change in the appearance of the vulvar mucous membrane. This latter effect was apparently due to alterations in vascularity, which in the early stages appeared as an intense engorgement, being followed by what appeared grossly to be a patchy thrombosis of some of the vessels; later an ischemia became apparent.

The Uteri: At autopsy it was possible, on the basis of their gross appearance, to arrange the uterine contents into four different groups: (1) the normal, in which the fetus, membranes, and placenta were obviously intact; (2) the necrotic, in which the placenta was a putty-like mass and the fetus and membranes had been discharged or were markedly autolyzed; (3) early abortion, in which the uterine segment was filled with a brownish, mucoid fluid, and the placenta and fetal structures showed some early histologic signs of destruction; (4) the hemorrhagic placenta, in which this structure was greatly infiltrated with blood (Table I).

Histologically signs of three apparently related pathological features of the uterine tissues were observed: namely (1) a necrosis of the fetal parts of the placenta; (2) an infarction of the placental blood vessels and a diapedesis and infiltration of red blood cells into the placental tissue; (3) an infiltration of leukocytes and round cells. The most advanced examples of the abortifacient effect showed an extensive necrosis of the placenta, but this did not include the deepest layer of the decidua, which was intact. The necrosis of the fetal parts involved the syncytial and Langhans' cells, both of which became highly vacuolated before they disintegrated. The endometrium elsewhere in the segment of the uterus was entirely normal.

The earlier examples showed both a fetal necrosis—i.e., vacuolation and disintegration of syncytial, Langhans', and membrane cells—and a thrombosis of the blood vessels, making it impossible to say which was primary. The infarction of the vessels extended to the deepest layer of the placenta. The polymorphonuclear and round-cell infiltration appeared to be secondary to the necrosis, and was usually limited to the periphery of such areas when they were large.

In the study of the microscopic characteristics of the placentae an attempt was made to discern various types of the abortifacient effect. The conclusion
was reached that the process was essentially the same in each aborted specimen, the variables being chiefly the intensity and duration of the effect.

The Ovaries: These organs appeared grossly normal and save in a few instances histological studies revealed no definite abnormalities in either follicles or corpora lutea. The exceptions were as follows: the ovaries of 3 rabbits injected with urine from cancer cases had a few miliary abscesses; the ovaries of 5 rabbits had in the corpora lutea what was interpreted as a degenerative effect, characterized by a foamy change in the luteal cells in patchy areas, in the center of which were accumulations of fibroblasts and polymorphonuclear cells. These latter changes were in no way similar to the miliary abscesses noted above. Each of the animals had aborted, 4 having been injected with urine and one with serum from cancer patients. This degenerative change could not be correlated with the degree of necrosis of the placenta, for certain animals with equally marked placental necrosis did not show this change.

The Kidneys: Approximately one injected animal in four had what were considered to be normal kidneys. The others had lesions which varied from grossly visible infarcts (4 animals) to microscopic accumulations of lymphocytes and polymorphonuclear leukocytes. One animal had what appeared to be a degenerative reaction in the glomeruli. These effects could not be correlated with cancer or non-cancer cases, with the injection of serum or urine, with aborting or normal uteri.

The Spleens: Many of the spleens were grossly swollen, congested, and friable, and most of those that appeared grossly normal showed microscopic evidence of disturbances characterized by congestion, hemorrhage, and necrosis. These changes, like those of the kidneys, could not be correlated with other factors. They probably were an expression of the animals' reactions to the rather drastic treatment to which they had been subjected.

DISCUSSION

These experiments demonstrated that the majority of the tested patients with cancer had in their serum and urine an agent which, when injected intravenously in pregnant rabbits, produced a necrosis of the fetal parts and also a thrombosis and hemorrhage in the placenta. Half of the tested non-cancer patients failed to show the presence of this agent in the urine or serum in the amounts injected. The phenomenon is not highly specific for cancer, since certain of the non-cancer patients did produce this agent and one of those with cancer did not. The abortifacient action occurred in a sufficient proportion of the cases, nevertheless, to justify more elaborate attempts to identify the agent and its mode of action.

On the basis of the experiments the following suggestions concerning the nature of the abortifacient action are offered:

(1) Extraneous causes such as trauma, fright, or spontaneous abortion do not appear to have been the cause of the abortions, save possibly in the 3 instances in which there was extensive hemorrhage into the placentae, where it is conceivable that trauma may have been a factor. Rabbits are recognized as being very tenacious of their pregnancies, and each of these animals had previously completed 2 normal pregnancies.
Although some endocrine imbalance as the result of the injections is a likelihood, the normal histologic appearance of the ovaries, which contained no freshly matured follicles and with a few exceptions no striking evidence of degeneration of the corpora lutea, gives no support for the belief that ovulation or a failure of the corpora lutea precipitated the abortions. Nor did a study of representative sections of the pituitaries by Dr. A. E. Severinghaus give any definite clue as to the possible rôle of this endocrine gland.

It seems probable that the effect was caused by some unknown toxic factor whose action was primary either upon the chorionic epithelium or upon the vascular structure of the placenta; the latter appearing the more likely supposition.

The occurrence of the abortifacient agent in the serum and urine of patients without demonstrable malignant disease seems to exclude the possibility that it is a unique product of cancer cells or of the host as a specific reaction to the presence of cancer. It must be, therefore, a principle that is produced by the host in response to conditions which are as yet unknown. The physician knows that advanced malignant disease is a malady with many facets, only one of which is the tumor itself, the reactions of the host to the tumor comprising the others. It is conceivable that the abortifacient herein discussed is produced by the body in response to a state which occurs in maladies other than cancer.

An interpretation of the results is made extremely difficult by our knowledge that the varied factors which lead to the early termination of pregnancy include trauma, drugs, infections, and disturbances of the serological, endocrine, and psychic balance in the body. Nor is much aid in the task forthcoming from the fact that, although malignant cells and chorionic or embryonal cells have certain features in common, they also have so many fundamental differences in morphology and physiology that one is forced to be skeptical that an agent produced by the one would naturally react specifically on the other.

These briefly mentioned complexities of the problem, instead of being obstacles, may serve as guides in further experiments which naturally suggest themselves somewhat as follows:

1. An attempt should be made to obtain the active agent in an extract or simple fraction of the urine so that its physiological and chemical properties may be studied apart from other contaminants.

2. Normal elderly patients without malignant disease, and those with other chronic and possibly acute illnesses should be tested for the abortifacient.

3. Patients with well-developed malignant tumors but without any signs of cachexia or weakness should also be tested for the presence of the agent in question.

**SUMMARY**

The experiments of Elsasser and Wallace, dealing with an abortifacient agent in the serum and urine of patients with cancer, have been repeated in part. The urine or serum of the majority of the tested cancer subjects contained a principle which caused the termination of early pregnancy in rabbits. The principle was not specific for cancer alone, because 3 of 6 apparently non-
cancerous patients had this agent in the blood or urine, and one of the cancer subjects did not have it in either serum or urine.

The principal lesions of the injected animals were apparent in the uteri, where there was a degeneration of the fetal structures, including the syncytial and Langhans' cells, a thrombosis of and hemorrhage from the placental vascular structure, and later in the process an infiltration of the necrotic tissue with leukocytes.

Note: The authors acknowledge the aid generously given by Dr. Stanhope Bayne-Jones, Dr. M. C. Winternitz, and Dr. A. E. Severinghaus.

To Dr. Theodore H. Elsasser goes the warm gratitude of the authors for giving so freely of his time to demonstrate his methods.