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BACTERIAL INTERFERENCE IN THE THERAPY OF RECURRENT STAPHYLOCOCCAL INFECTIONS*

Multiple Abscesses Due to the Implantation of the 502A Strain of Staphylococcus

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BACTERIAL interference has been successful in curtailing epidemics of staphylococcal disease in the nursery.¹⁻⁴ Implantation of a relatively non-pathogenic strain of *Staphylococcus aureus* on the nasal mucosa and umbilical stumps of newborn infants has prevented colonization with epidemic staphylococcal strains, thus interrupting their transmission from infant to infant. A strain of *Staph. aureus* designated 502A has been employed in these studies. Persistent nasal carriage of pathogenic staphylococci in adults has also been interrupted by implantation of 502A on the nasal mucosa after antibiotic suppression of the resident flora.^{5,6} To date no cases of serious staphylococcal disease due to this micro-organism have been noted after its implantation.⁷

In patients with recurrent staphylococcal skin disease there is some evidence that pathogenic staphylococci are seeded to the skin from the nasal mucosa.⁸ Thus, replacement of the pathogenic staphylococcus residing in the nose by a strain of minimal pathogenicity such as 502A might lower the incidence of skin infections. We have recently studied a patient with a chronic skin disorder upon which were superimposed frequent, deep-seated, staphylococcal skin abscesses. Because she was a nasal carrier of a staphylococcal strain apparently identical to that recovered from her skin lesions, replacement of her resident nasal staphylococci with the 502A strain was attempted. Although induction of the 502A nasal-carrier was associated with a marked decrease in the incidence of skin abscesses, the 502A strain was isolated from all but 1 of the abscesses that subsequently developed. In 2 cases 502A was isolated in pure culture. This is the first time, to our knowledge, that this micro-organism has been implicated in the production of frank abscesses.

CASE REPORT

B.E. (V.U.H. 139418), a 21-year-old woman, was in excellent health until late 1955, when at the age of 11, a pruritic nodular skin eruption on the face, neck and extremities

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developed. These skin lesions of unknown etiology persisted, recurring in crops over the ensuing years. At the age of 18 she was hospitalized briefly for pneumonia and was treated with penicillin. A few days after discharge the 1st skin infection, a draining pustular calf lesion that resolved without therapy developed. Three months later another calf abscess appeared, and abscesses on the legs subsequently recurred at intervals of several months. There was no family history of dermatologic disease and she had no known exposure to patients with staphylococcal infections.

In October, 1963, at the age of 19, she was admitted to Vanderbilt University Hospital for evaluation of the underlying chronic skin disorder. At that time examination revealed well circumscribed, erythematous, round to oval nodules ranging from 0.5 to 1.5 cm. in diameter and 2 to 3 mm. in height, localized primarily to the extremities but also present on the upper trunk, neck and face. The lesions were excoriated and crusted, and the intervening skin was thick, dry and lichenified. No skin abscesses were present. An extensive investigation, including 3 skin biopsies, failed to elucidate the nature of the skin disease.

The course during the 22-month period from December, 1963, to September, 1965, is summarized in Figure 1. Forty staphylococcal abscesses were documented over this time. The abscesses were limited to the lower extremities in the areas where the underlying skin disease was the most severe and the trauma of scratching the most pronounced.

In May, 1964, the patient was instructed to begin washing the skin regularly with hexachlorophene (pHisoHex). She also was treated intermittently with oxacillin by mouth until June, 1965, when therapy with penicillin V and intranasal

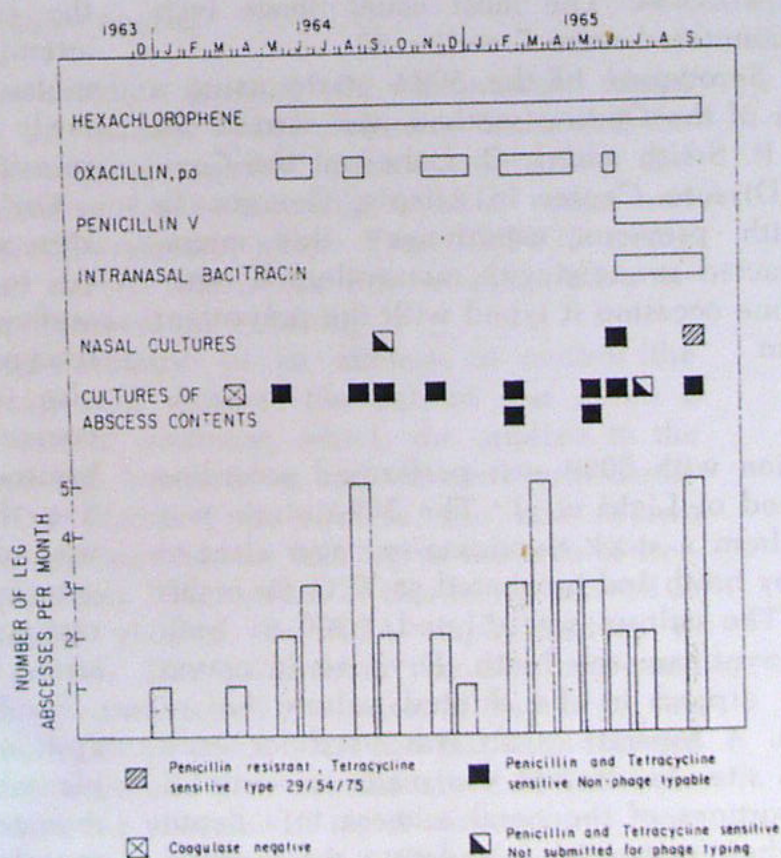


FIGURE 1. Clinical Course of the Patient before the Initiation of the 502A Implantation Program.

bacitracin ointment was begun. These agents failed to reduce the incidence of the abscesses.

Of the 40 abscesses 12 were cultured. In 10, coagulase-positive nonphage-typable staphylococci, sensitive to penicillin and to tetracycline, and also identical in the remainder of their antibiotograms, were recovered. Two nasal cultures revealed coagulase-positive staphylococci with this same antibiotic sensitivity pattern. The only strain submitted for phage typing was nontypable. The skin and nasal staphylococci were apparently identical. In September, 1965, after a 14-week period of penicillin V therapy, a nasal culture revealed a penicillin-resistant, coagulase-positive staphylococcus for the 1st time. This micro-organism was sensitive to tetracycline and was lysed by phages 29/54/75.

The failure of antibiotic therapy and attention to skin hygiene to benefit this patient suggested bacterial interference as a mode of treatment.

MATERIALS AND METHODS

The Organism

The 502A strain of staphylococcus was obtained from Dr. John C. Ribble, of the New York Hospital-Cornell Medical Center. Throughout this study the identification of this coagulase-positive micro-organism was based on 3 sets of characteristics.

Antibiogram. 502A was sensitive to the following antibiotic disks: penicillin, 2 units; erythromycin, 2 μ g.; kanamycin, 5 μ g.; novobiocin, 5 μ g.; and oxacillin, 1 μ g. The strain of 502A utilized was resistant to both 5 μ g. and 10- μ g. tetracycline disks. The original 502A strain utilized by Shinefield and his associates⁹ was reported to be sensitive to 10- μ g. tetracycline disks, but for some time this strain has been noted to be resistant to tetracycline disks of both strengths.¹⁰

Phage type. At routine test dilutions there was lysis by one or more of the following group III phages: 6/7/47/53/54. The most usual phage lysis patterns encountered were 7 and/or 53.

Serotype. Serotyping of the 502A strain using a modification of the Oeding method was carried out by Drs. P. B. Smith and J. O. Cohen at the Communicable Disease Center in Atlanta, Georgia. In keeping with previous experience¹¹ this micro-organism reacted strongly with monovalent C₁ antiserum. On one occasion it typed with the polyvalent Cp antiserum.

Inoculation

Colonization with 502A was performed according to the method of Light et al.⁴ The 502A strain was transferred from a stock trypticase-soy agar slant to trypticase soy broth and incubated at 37°C. for eighteen hours. The culture was diluted 1:200 in 1-ml. aliquots of trypticase soy broth. Dry, sterile cotton swabs were dipped in the diluted culture before implantation. A separate swab was used for each implantation site. An attempt was made to reach all accessible portions of the nasal mucosa to a depth of 2 cm. when intranasal inoculation was carried out. 502A implantation was performed every second or third day for 4 applications; subsequent implantations followed at weekly intervals.

Recovery and Identification

Dry, sterile cotton swabs were rotated several times in each nostril to a depth of 2 cm. Cultures of the axilla, perineum and skin surfaces were made by "scrubbing" of these areas with a dry cotton swab. Thorough cleansing of the skin preceded incision and drainage of abscesses, but spontaneously draining lesions were cultured directly without cleaning of the skin first.

All swabs were streaked directly on mannitol-salt agar plates, which were incubated for thirty-six hours at 37°C. The colonies were then examined for morphology, pigmentation and fermentation patterns. Large, opaque colonies were always selected in preference to salt-inhibited pinpoint colonies. A minimum of 5 such colonies per plate was selected for further study. Each colony was tested for coagulase production with pooled human plasma. Antibiotic-sensitivity patterns were determined on sheep-blood agar plates with the use of the disk concentrations noted above. Throughout the period of study representative coagulase-positive staphylococci were submitted to the Communicable Disease Center in Atlanta, Georgia for phage typing and serotyping.

THE STUDY

The details of the implantation study are shown in Figure 2. Early in October, 1965, selected survey sites (both nostrils, left axilla, skin of left forearm and perineum) were cultured to determine the patient's prestudy staphylococcal flora. Oxacillin, 6 gm. daily by mouth, and intranasal instillation of bacitracin ointment were started. Despite this therapy the staphylococcal flora was not changed, and 4 attempts to establish the 502A strain on her nasal mucosa were unsuccessful. The study was temporarily interrupted and hexachlorophene scrubs were reinstated.

Early in November cultures of the original survey sites showed the phage Type 29/54/75 staphylococcus to be in the nose, on the perineum and on the surface of a nondraining abscess on the left leg. The tetracycline-sensitive nonphage-typable strain was also isolated from the latter 2 sites. A two-week course of therapy with cloxacillin, 4.5 gm. daily, and bacitracin nasal ointment was begun. Over the next five days 2 leg abscesses developed. Probenecid was added to the regimen, but despite a cloxacillin serum level of 25 μ g. per milliliter, the penicillin-resistant, phage Type 29/54/75 staphylococcus persisted in the nose.

Six hours after termination of antimicrobial therapy on November 24, all survey sites were implanted with 502A. Nasal implantation of 502A was repeated two and five days later. Cultures initially revealed persistence of the patient's resident staphylococcal flora, but cultures obtained on December 2 revealed that 502A had been established on the nasal mucosa in the continuing presence of the

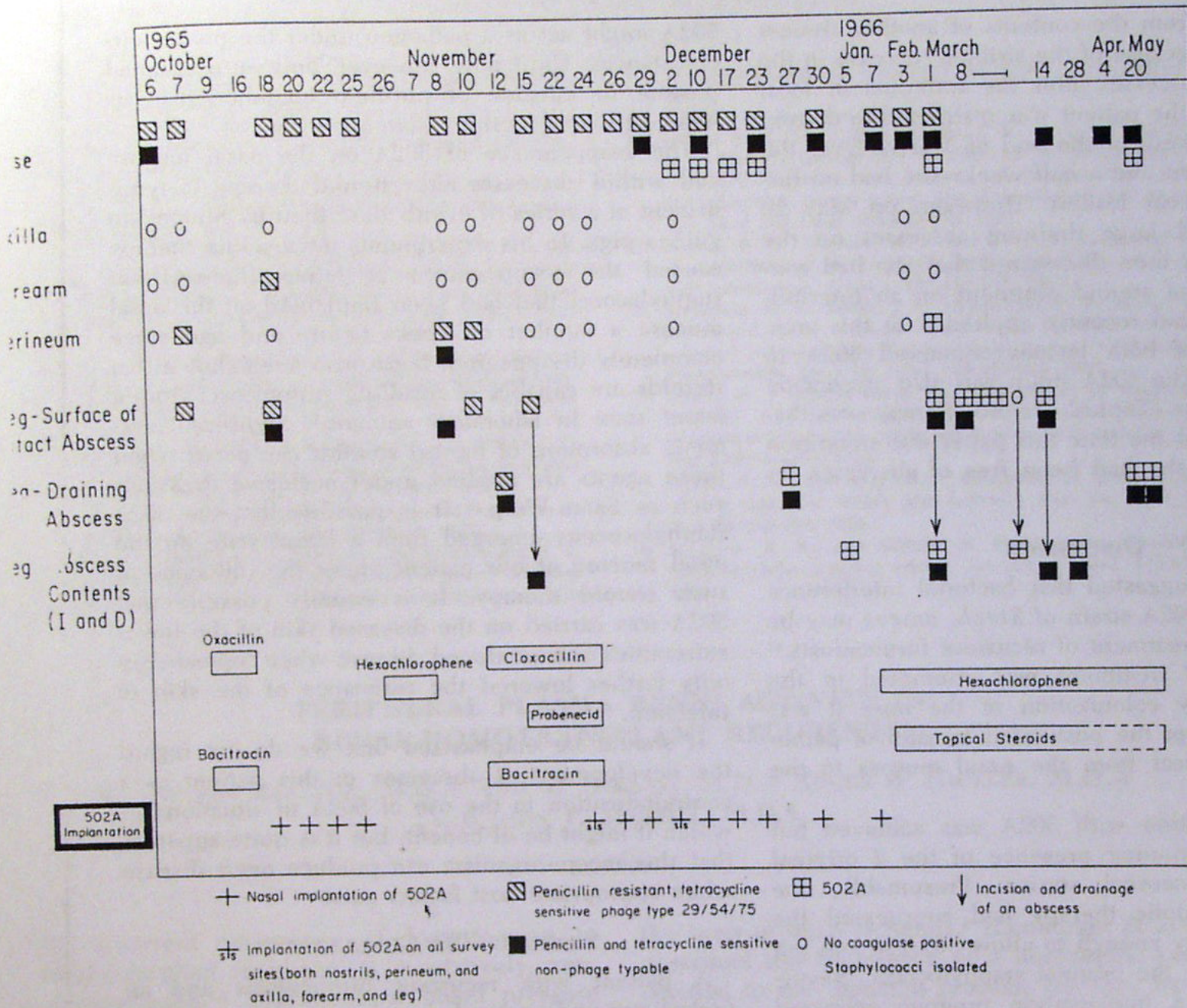


FIGURE 2. Course of the 502A Implantation Program.

...e Type 29/54/75 strain. No other sites yielded ase-positive staphylococci. Subsequent cul- s: weekly intervals each disclosed 502A on the d mucosa along with the penicillin-resistant ge Type 29/54/75 strain. The nontypable strain was sensitive to penicillin and tetracycline also eared in the nose on two occasions. n December 27 a draining abscess was observed the left calf. This was the first such lesion in y-two days and occurred thirty-three days after last dose of cloxacillin. Culture of the drainage ded the 502A staphylococcus and the non-ge-typable strain sensitive to penicillin and tet- cline. Twenty-five days had elapsed since 502A last been implanted on the skin, but it had n continuously present in the nose. n December 30, for the first time in five weeks, nasal culture before implantation failed to reveal A. Six days after this last nasal 502A implanta- the patient presented a very large abscess on right leg. The lesion was widely incised after eful surgical preparation. Culture of the abscess tentents revealed a pure growth of 502A. When the ults of this abscess culture became apparent the

502A implantation program was discontinued. Nasal cultures on January 7 and February 3 showed the patient's original resident penicillin-sensitive and tetracycline-sensitive nonphage-typable strain and the penicillin-resistant phage Type 29/54/75 strain. All other survey sites were free of coagulase-positive staphylococci on February 3. In mid-February, in an attempt to control the underlying skin disease, the patient was given a topical steroid ointment, which she applied to the legs from the knees down and covered with Saran-Wrap. On March 1 an abscess, the first in two months, appeared on the left leg in the area of steroid application. The surface of this lesion as well as its contents yielded the 502A staphylococcus in mixed culture. The 502A strain was also recovered from the perineum and the nose. It had been two and a half months since 502A had last been isolated from the nose and two months since it had been found in a leg abscess. In rapid succession 4 more abscesses appeared on the leg. One of these lesions was incised and drained, revealing 502A in pure culture. This micro-organism was also recovered in mixed culture from the surface of an abscess on

March 14 and from the contents of another abscess on March 28. Because of the striking increase in the incidence of abscesses after the initiation of local steroid therapy the patient was instructed to discontinue this treatment at the end of March. Over the subsequent seven and a half weeks she had no further staphylococcal lesions. However, on May 20 she presented 2 large draining abscesses on the right leg. It was then discovered that she had continued the use of steroid ointment on an intermittent basis and had recently applied it to this area. The drainage of both lesions contained 502A in mixed culture. The 502A strain was also present on the nasal mucosa. Topical steroid therapy was discontinued, and at the time this paper was submitted for publication, she had been free of abscesses for one month.

DISCUSSION

It has been suggested that bacterial interference employing the 502A strain of *Staph. aureus* may be utilized in the treatment of recurrent furunculosis.¹² This method of treatment was attempted in the present case. By colonization of the nose it was hoped to interrupt the postulated transfer of pathogenic staphylococci from the nasal mucosa to the skin.

Nasal colonization with 502A was achieved but only in the continuing presence of the 2 original resident staphylococcal strains. Presumably, the antecedent antibiotic therapy had suppressed the resident flora only enough to allow 502A to join but not to supersede the original staphylococci. Nevertheless, the 502A implantation program appeared in some way to have interrupted the recurrent episodes of furunculosis in this patient. It seems unlikely that a natural fluctuation in the activity of the disease was responsible; the patient could not remember spontaneous freedom from abscesses in some thirty months. It is also doubtful if the antibiotic therapy administered in conjunction with the 502A implantation program lowered the incidence of abscesses since several previous courses of anti-staphylococcal antibiotic therapy were without benefit. Perhaps the presence of 502A on the nasal mucosa served to suppress the numbers of the 2 more pathogenic strains, thus resulting in delivery of fewer virulent micro-organisms to the skin. This might represent a state of "partial" bacterial interference rather than the complete bacterial interference described by Shinefield and his colleagues.⁹

The development of an abscess clearly due to the 502A strain of *Staph. aureus* demonstrates that this micro-organism can produce overt disease. It is likely that the patient's underlying skin disease made her unusually susceptible to progressive infection in an undefined way. This once again emphasizes the role of the host in determining the disease-producing potential of any micro-organism. Shinefield and his co-workers¹³ anticipated that

502A might act as a pathogen under the proper circumstances. Until now, however, only an occasional pustule or episode of purulent conjunctivitis has been attributed to this micro-organism.⁷

The reappearance of 502A on the nasal mucosa and within abscesses after steroid therapy is reminiscent of a series of events described by Simon¹⁴ in guinea pigs. In his experiments tetracycline therapy caused the reappearance of tetracycline-resistant staphylococci that had been implanted on the nasal mucosa a number of weeks before and had since completely disappeared. It has also been shown that steroids are capable of recalling streptococci from a latent state in laboratory animals.¹⁵ Significant systemic absorption of topical steroids can occur when these agents are applied under occlusive dressings such as Saran-Wrap.¹⁶ It is possible that the 502A staphylococcus emerged from a latent state on the nasal mucosa of our patient under the influence of such steroid therapy. It is equally possible that 502A was carried on the diseased skin of the lower extremities and produced disease when topical steroids further lowered the resistance of the skin to infection.

It should be emphasized that we do not regard the development of abscesses in this patient as a contraindication to the use of 502A in situations in which it might be of benefit, but it is quite apparent that this micro-organism can produce overt disease when appropriate host factors exist.

SUMMARY AND CONCLUSIONS

A patient with recurrent furunculosis and an underlying undiagnosed skin disease was treated by means of bacterial interference utilizing the 502A strain of *Staphylococcus aureus*. The nasal mucosa was successfully colonized with 502A although elimination of the original staphylococcal flora was not achieved. The incidence of staphylococcal lesions was reduced on this program. However, 502A was isolated from all but 1 of the abscesses that did occur subsequently, twice in pure culture. This is the first reported case of staphylococcal disease that followed the implantation of the 502A staphylococcus. This experience does not constitute a contraindication to the use of the 502A strain when it may be of benefit, but it is apparent that this "relatively nonpathogenic micro-organism" can produce staphylococcal disease when appropriate host factors exist.

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PERIPHERAL PLASMA RENIN ACTIVITY IN RENAL-HOMOTRANSPLANT RECIPIENTS*

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BOSTON

THE transient occurrence of hypertension in renal-transplant recipients is a relatively common phenomenon.^{1,2} Elevations of blood pressure levels higher than 140 systolic, 90 diastolic) usually occur within a few days after insertion of the graft and seldom persist permanently.³ Most investigators have noted a clear relation between the clinical rejection of the kidney, salt retention, weight gain, decreasing renal function and hypertension.⁴ The diastolic blood pressure often reaches levels exceeding 120 mm. of mercury, convulsions and other signs of encephalopathy uncommon.

The possible etiologic factors that have been implicated in the rise in blood pressure in recipients transplants include salt retention, with secondary expansion of the intravascular space,⁵ and decreased renal blood flow, with subsequent hypertension on

the basis of renal ischemia.⁶ Hamburger et al.⁷ have described the occurrence of a nonspecific systemic reaction to the rejection process, but they have not noted hypertension.

A recent report describes a case of hypertension in a renal-transplant recipient in which elevations of peripheral plasma renin activity were noted⁸ during the rejection crisis. Shibagaki and his co-workers⁹ have noted a decrease in the renin content of the kidneys of patients receiving transplants. The present study was designed to investigate the possible implication of the renin-angiotensin system in the hypertension associated with renal transplantation.

METHODS

Peripheral plasma renin activity was measured in 11 patients with terminal renal failure supported by chronic hemodialysis while they were awaiting transplantation. In 3 of these 11 patients the assays were repeated after bilateral nephrectomy. Serial renin assays were performed on 7 after renal transplantation. Five of the subjects received kidneys from live related donors, 1 received a kidney from an unrelated donor, and 1 received the graft from a cadaver. Twenty-seven renin measurements were performed after transplantation. Five were done on the first day after transplantation, and the rest at intervals during the first three weeks.

The renin was assayed by a modification of the method of Boucher et al.¹⁰ The plasma samples

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