MINIREVIEW

Serum Therapy Revisited: Animal Models of Infection and Development of Passive Antibody Therapy

ARTURO CASADEVALL1,2* AND MATTHEW D. SCHARFF3

Division of Infectious Diseases, Department of Medicine,1 and Departments of Microbiology and Immunology2 and Cell Biology,3 Albert Einstein College of Medicine, Bronx, New York 10461

INTRODUCTION

In the preantibiotic era, passively administered immune animal sera, or serum therapy, was the primary mode of treatment for many infectious diseases, including diphtheria, tetanus, scarlet fever, pneumococcal pneumonia, and meningitis caused by Neisseria meningitidis and Haemophilus influenzae (24). Immune sera contained specific antibodies which mediated therapeutic effects by promoting opsonization, neutralizing toxins, and/or triggering complement-mediated bacterial lysis. Toxicity resulting from the systemic administration of foreign proteins was associated with serum therapy, however, and so serum therapy was abandoned when antibiotics became widely available in the 1940s.

Over the past half-century there has been relatively little interest in passive antibody therapy for bacterial and fungal infections because effective antimicrobial drugs have been available. However, several recent developments should renew interest in the use of passive antibody therapy alone or in combination with antimicrobial drugs. First, the emergence of antimicrobial resistance has decreased the efficacy and predictability of antimicrobial chemotherapy. Second, the difficulties of treating infections in immunocompromised individuals, particularly those with AIDS, have revealed the limitations of antimicrobial chemotherapy in the absence of effective immunity. Third, the hybridoma technology introduced in 1975 by Kohler and Milstein (68) provides the means of generating an unlimited supply of homogeneous monoclonal antibodies (MAbs). Technology is now available to reduce the immunogenicity of rodent MAbs in humans by constructing mouse-human chimeric or humanized MAbs (77) or to generate completely human MAbs from either hybridomas or combinatorial libraries (65). Thus, antibody-based therapies no longer depend on heterologous immune sera, with their inherent variations and toxicities, and antibodies can again be considered therapeutic alternatives for a variety of infections.

Potentially useful antibodies for the prevention and therapy of infectious diseases are usually identified by demonstrating that they can modify the course of experimental infection. The choice of an animal model for use in the testing of antibody reagents can be a critical decision for demonstrating efficacy. The development of serum therapy in the preantibiotic era relied almost exclusively on animal models in the preclinical testing phase. Here we review serum therapy for pneumococcal and meningococcal infections, with emphasis on the role of the animal models used in their development.

SERUM THERAPY FOR STREPTOCOCCUS PNEUMONIAE INFECTIONS

Development. Streptococcus pneumoniae (pneumococcus) is an encapsulated gram-positive diplococcus which causes lobar pneumonia. The mortality rate from untreated pneumococcal pneumonia is 20 to 40% (Table 1). The potential usefulness of passively administered antibody for treating pneumococcal infection was first shown in 1891 when the Klemperers protected rabbits with immune serum (67). In 1914, Cole (26) stated that pneumococcal pneumonia was the most important infectious disease of his time, and the development of antipneumococcal sera received considerable attention.

Antipneumococcal sera for human therapy were identified by their ability to protect experimental animals against lethal infection. The most important system for testing antipneumococcal sera was the mouse model of intraperitoneal (i.p.) infection (32, 34), in which both the infection and the serum were given i.p. at the same time and often after being mixed in one syringe (55, 84). Neufeld and Haendel (80, 81), Avery (2), Dochez (32), and Dochez and Gillespie (34) used the mouse i.p. model to demonstrate that only type-specific sera were protective. Dochez (32) concluded from mouse protection data that sera that were not protective in mice would not be protective in humans, and the protective efficacies of sera were correlated with their agglutinating capacities (26, 32, 34). The mouse i.p. model was also used to demonstrate the presence of antibodies in the convalescent-phase serum of patients with pneumococcal pneumonia (31, 70) and to determine the efficacy of antipneumococcal vaccines in inducing protective antibodies (44). The advantages of the mouse model were that mice were very susceptible to pneumococci, mice were a readily available laboratory species, and more importantly, the model discriminated between protective and nonprotective sera.

The potency of antipneumococcal sera was standardized in the mouse i.p. model (35, 36). A unit of potency was defined as 10 times the smallest amount of serum which protected two of three mice from a “hundred thousand minimal fatal doses of very virulent pneumococci” (because of individual mouse variation, the criterion of 100% survival was considered too stringent) (84). The mouse test, although useful, was far from ideal. Individual variation required the use of a minimum of 10 mice for each dilution of serum, which contributed a significant expense to serum preparation (37). Despite standardization, the therapeutic efficacies of different serum preparations containing the same number of units were not necessarily equivalent because of differences in virulence between the pneumococcal strains used for standardization and clinical strains (28). In fact, the reproducibility of mouse protection test results within a laboratory that used standard strains was often poor...
The mouse protection test also predicted a "prozone" effect, in which no protection would occur if large amounts of immune sera were used (35, 55). The prozone effect in mice was believed to be an in vivo equivalent of prozone effects in in vitro precipitation and agglutination reactions (55). However, a prozone effect was not observed in humans given large amounts of serum (16, 56) or in mice given large amounts of rabbit serum (60) and possibly represented a poorly understood artifact of the effect of horse serum on the mouse model which was not relevant to clinical practice. The difficulties associated with the mouse protection test led to attempts to standardize sera by using precipitation of pneumococcal antigens and agglutination (37, 99), but none of these techniques achieved general acceptance (98).

Other animals used in the development of antipneumococcal therapy were monkeys and rabbits. Cecil and colleagues (19, 23) used a monkey (Macacus sinicu) model in which pneumococcal pneumonia was induced by intratracheal inoculation of S. pneumoniae to confirm that only type-specific sera were protective and to justify the rationale for the serum therapy of pneumococcal pneumonia. In rabbits, subcutaneous inoculation of S. pneumoniae produced disseminated infection, with >80% mortality and pulmonary findings of pneumonia (53). The rabbit dermal pneumonia model was used to demonstrate the efficacy of passive antibody against pneumococcus (53) and to estimate the appropriate dose of serum for use in humans (54).

Antipneumococcal sera were made in a variety of animals, with horses and rabbits being most frequently used (27, 61). Type-specific horse antipneumococcal sera were generated by complicated immunization protocols which used both dead and live bacteria (27). The efficacy of immunization was determined by the power of serum to agglutinate pneumococci and protect mice (27). Production of antipneumococcal serum in horses required months, and the costs involved in the veterinary care of the animals, purification of antibody, and testing in mice made serum therapy expensive. Rabbit sera offered certain advantages over horse sera, including higher specific activity, antibodies with lower molecular weights (which were claimed to have higher levels of tissue penetration), no prozone effects in the mouse model, more rapid immunization protocols, and reduced cost (60, 61).

**Clinical use.** Serum therapy was most effective if it was begun within 3 days of the onset of pneumococcal pneumonia (23, 42). The mortality of type I pneumonia could be reduced to 5% by administration of serum within the first 24 h of the onset of symptoms (18). Administration of serum 4 to 5 days after symptoms began was believed to produce little benefit (16, 43). To avoid delays associated with the recovery and typing of clinical isolates, serum therapy was often begun empirically by using polyvalent preparations generated by mixing monovalent sera (15, 42). The mouse i.p. model was used in clinical practice to recover pneumococcal isolates for typing (20, 26, 90). The recovery of clinical isolates involved injecting the patient's blood and/or sputum i.p. into a mouse, waiting 4 to 5 h, and then washing the peritoneal cavity of the mouse with a salt solution to obtain a suspension of pneumococci for typing (26). When a patient responded poorly to type-specific therapy and a mixed infection was suspected, mice could be used to isolate other pneumococcal types that were present (16). This was done by injecting the patient's sputum into two mice, one of which was also given the serum used in the initial therapy. The mouse receiving only sputum would develop bacteremia of the original pneumococcal type, whereas that strain would be suppressed by the serum in the second mouse. If a mixed infection was present, the second mouse would develop bacteremia caused by the occult serotype (42), which could then be typed. Serum was usually administered to patients intravenously (i.v.) after testing for hypersensitivity reactions by injecting a small dose subcutaneously or instilling a dilute serum solution into the conjunctival sac (16, 23, 43, 56). The intramuscular (29, 84) and subcutaneous (17) routes were sometimes used to reduce the likelihood of immediate reactions (chills, anaphylaxis), but i.v. administration was preferred because it was the most efficient in achieving high antibody concentrations in

### Table 1. Efficacy of serum therapy for pneumococcal pneumonia

<table>
<thead>
<tr>
<th>Author(s)</th>
<th>Year</th>
<th>Reference</th>
<th>Serum treated*</th>
<th>Non-serum treated*</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cecil and Larsen</td>
<td>1922</td>
<td>20</td>
<td>331</td>
<td>62</td>
<td>18.7</td>
</tr>
<tr>
<td>Cecil</td>
<td>1925</td>
<td>17</td>
<td>73</td>
<td>14</td>
<td>19.2</td>
</tr>
<tr>
<td>Cecil and Slufft</td>
<td>1928</td>
<td>23</td>
<td>401</td>
<td>116</td>
<td>28.9</td>
</tr>
<tr>
<td>Rosenbluth</td>
<td>1928</td>
<td>89</td>
<td>210</td>
<td>40</td>
<td>19.0</td>
</tr>
<tr>
<td>Bulkow</td>
<td>1928</td>
<td>13</td>
<td>135</td>
<td>23</td>
<td>17.0</td>
</tr>
<tr>
<td>Park et al.</td>
<td>1928</td>
<td>83</td>
<td>156</td>
<td>31</td>
<td>19.9</td>
</tr>
<tr>
<td>Cecil and Plummer</td>
<td>1930</td>
<td>21</td>
<td>102</td>
<td>12</td>
<td>11.6</td>
</tr>
<tr>
<td>Finland</td>
<td>1930</td>
<td>42</td>
<td>203</td>
<td>45</td>
<td>22.1</td>
</tr>
<tr>
<td>MacCordick</td>
<td>1931</td>
<td>71</td>
<td>77</td>
<td>7</td>
<td>9.1</td>
</tr>
<tr>
<td>Bulkow</td>
<td>1933</td>
<td>14</td>
<td>385</td>
<td>72</td>
<td>18.7</td>
</tr>
<tr>
<td>Finland and Slufft</td>
<td>1933</td>
<td>45</td>
<td>46</td>
<td>9</td>
<td>19.6</td>
</tr>
<tr>
<td>Medical Research Council</td>
<td>1934</td>
<td>72</td>
<td>184</td>
<td>18</td>
<td>9.7</td>
</tr>
<tr>
<td>Belk</td>
<td>1935</td>
<td>46</td>
<td>463</td>
<td>75</td>
<td>16.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td><strong>1,815</strong></td>
<td><strong>374</strong></td>
<td><strong>20.6</strong></td>
</tr>
</tbody>
</table>

* Patients with type III pneumococcal pneumonia were excluded because no effective serum against this serotype was ever developed. The numbers listed are for individuals with pneumonia and include bacteremic and nonbacteremic subjects.

P values were calculated by chi-square analysis by using Primer of Statistics: The Program (71a).

Study in which an attempt was made to use a case-control group.

In the study of Rosenbluth (89), there was a striking benefit for the subset of patients with pneumococcal bacteremia treated with serum (P < 0.001).

The study of Belk (4) is a retrospective statistical analysis of previous studies with serum in which he pooled published data, including data from some of the studies cited here. Belk included only those published studies for which there was sufficient data for treatment and control groups for analysis.

Data from Table 1 in reference 4.

Data from Table 2 in reference 4.

Downloaded from http://aac.asm.org/ on August 14, 2017 by guest
blood (94). Initial dosages varied with the study and the serum preparation used and ranged from 20,000 to 200,000 units per day (16, 21–23, 42, 56, 72, 84, 90). The dosage used was based on the severity of infection, the presence of bacteremia, the day of disease at which therapy was begun, and the response to initial therapy (4, 23, 55). Appropriate dosages were also estimated by measuring the agglutination titer of the patient’s serum after serum administration (16, 84, 90). The duration of treatment and total dosage were largely determined by the patient’s response to therapy, and benefits from antibody therapy were often evident within the first day of serum administration (16, 43, 72). Failure of serum therapy was associated with delayed administration, insufficient dosages, incorrect typing, the presence of mixed infections, the presence of abscesses, and overwhelming infection (16, 43, 56).

**Efficacy.** The efficacy of antipneumococcal sera remained uncertain until the discovery of pneumococcal serotypes and the need to use type-specific sera in therapy (25, 26, 80, 81). No effective sera for type III pneumococcal pneumonia was available (17, 43, 83). In the 1920s and 1930s several large studies showed that type-specific serum therapy decreased the rate of mortality from pneumococcal pneumonia (Table 1). Many of those studies were early examples of case-controlled studies in which the treatment and nontreatment groups were selected on the basis of either alternate admissions (13, 17, 42, 45, 72), hospital ward assignment (20), or chart number (23). However, those studies were not blinded and did not include a placebo control group. Establishment of the efficacy of serum therapy for pneumococcal pneumonia required studies with large numbers of subjects because the disease was often self-limited and many variables such as early timing of administration, accurate typing of isolates, and adequate dosage needed to be understood. Retrospective statistical analysis of the data strongly supports the conclusion that serum therapy was effective in reducing mortality (Table 1). By the late 1930s and early 1940s serum therapy for pneumococcal pneumonia was standard practice (16, 24, 56), and commercial type-specific sera were available for many of the pneumococcal types (Fig. 1).

The exact mechanism(s) by which antipneumococcal sera mediated therapeutic effects remains uncertain. Antipneumococcal sera had direct antibacterial effects (5, 33, 88) and promoted opsonization and agglutination of pneumococci in vivo (11, 12). Some studies described antitoxin properties for antipneumococcal sera (85, 91). In humans, antipneumococcal sera were effective in terminating bacteremias and limiting the extension of pulmonary consolidation (93). Immune antipneumococcal sera produced direct growth-inhibiting effects on pneumococci (5, 33, 88). Thus, the therapeutic efficacy of immune sera probably resulted from a combination of enhanced host immunity and direct antibacterial effects.

**SEUM THERAPY FOR NEISSERIA MENINGITIS INFECTIONS**

**Development.** Neisseria meningitidis ( meningococcus, also called Diplococcus intracellularis meningitides and Neisseria intracellularis in older literature [9]) is a gram-negative diplococcus which is the causative agent of epidemic meningitis (cerebrospinal fever). A pandemic in the early 1900s with a mortality rate of 70 to 80% provided a major impetus for the development of serum therapy for meningococcal meningitis. Serum therapy for meningococcal infections was developed by Jochmann (64) in Germany and Flexner (47, 49) and Flexner and Jobling (50) in the United States. Jochmann (64) demonstrated serum-mediated protection in mice and guinea pigs and treated patients, reporting a clinical benefit in 12 of 17 cases. Flexner (46, 48) studied meningococcal infection in several animals, but he primarily used monkeys (Macacus nemestrinus) in preclinical testing. He developed a monkey model with pathological features similar to those of human meningococcal meningitis by intraspinal (subarachnoid) inoculation (48). Administration of antimeningococcal serum directly into the subarachnoid space shortly after infection cured the majority of treated monkeys (46, 47, 97).

By the early 1930s empiric serum therapy was recommended for all children with presumed meningitis (79). However, the occurrence of several epidemics with high mortalities, despite serum treatment, renewed interest in the development of more effective sera (6, 8, 9, 82, 96). Unlike pneumococcal pneumonia (which in the murine i.p. model provided a fairly reliable system for the testing of serum reagents), the development of more effective antimeningococcal sera was hampered by difficulties with animal models (8, 9, 75). Mice and guinea pigs were resistant to infection, and the size of the inoculum required to kill them was so large that death was often attributed to toxicity rather than infection (7, 9). The mouse was not a reliable system for testing sera until Miller (74) and Miller and Castles (75) discovered that i.p. administration of mucin rendered mice susceptible to meningococcal strains. Mucin interfered with phagocytosis, and i.p. inoculation of meningococci resulted in lethal infection in mice which could be aborted with antimeningococcal sera (7, 75). Small inocula (as few as 10 bacteria) were lethal for mucin-treated mice (75). In addition to difficulties with animal models, the development of antimeningococcal sera was complicated by the fact that strains usually lost their virulence in vitro (9). In this regard, mucin-treated mice also provided a system for maintaining strain virulence by frequent passage in mice (9).

Antimeningococcal serum was a polyvalent solution generated by immunizing horses intravenously or subcutaneously with multiple live strains and bacterial lysates (37, 58, 66). Some immunization protocols used up to 50 different strains (58). A different approach was the development of meningococcal antitoxin generated by immunizing horses with meningococcal culture filtrates. The antitoxin strategy was based on the observation that culture filtrates could induce pathological lesions similar to those observed in meningococcal infection (38–40). However, the existence of a true meningococcal exotoxin was controversial, and some argued that the filtrate toxin was endotoxin released by bacterial autolysis (51). Antitoxin was tested in a variety of animal models, including guinea pigs, rabbits, and monkeys (38, 39, 41), was found to be protective, and was used in clinical practice, reported with good results (63). Comparison of antitoxin and antimeningococcal serum in mucin-treated mice suggested that the latter was more effective (66).

**Clinical use.** The high rate of mortality from human meningococcal infections combined with the urgency brought on by the epidemic in New York City in 1905 and 1906 resulted in the rapid clinical use of the antiserum of Flexner and Jobling (50). On the basis of their experience with the monkey model, human infections were treated with lumbar subarachnoid injections of horse sera (50). The injection protocol involved lumbar puncture, removal of a volume of cerebrospinal fluid greater than the intended dose of serum, and slow introduction of 30 ml of antimeningococcal serum by gravity flow (50, 78). Dosing was repeated every 24 h until the patient improved (78). If sepsis was suspected, the patient was also given serum by the i.v. route. Interestingly, intrathecal serum administration was thought to be safer than administration by the i.v. route, given that the latter carried a small risk of anaphylaxis...
The Life Saving Value of
ANTIPNEUMOCOCCIC SERUM · TYPE I

The value of this antibody concentrate in the treatment of Type I lobar pneumonia is clearly indicated by taking the totals of figures reported during the past 4 years by PARK, BULLOWA and RONEN-BLUTH; BALDWIN; CREEL & PLUMMER; FINLAND; HEPFORD; LORD; SUTLIFF; and CRASTER.

When the serum-treated cases are restricted to those treated within 72 hours after onset, there is a further reduction in mortality as follows—

<table>
<thead>
<tr>
<th>Case</th>
<th>Doses</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td>553</td>
<td>166</td>
</tr>
<tr>
<td>Treated</td>
<td>707</td>
<td>131</td>
</tr>
</tbody>
</table>

It is estimated that of approximately 30,000 deaths in the United States every year from Type I pneumonia, two-thirds could be avoided by the early administration of Type I Serum.

From the standpoint of lower mortality as well as economy of treatment, Antipneumococcic Serum Type I (Lederle) deserves your consideration

Literture upon Request

LEDERLE LABORATORIES, INC., NEW YORK

FIG. 1. Advertisement for serum to type I pneumococci made by Lederle Laboratories which appeared in the February 1933 issue of the New York State Journal of Medicine (Vol. 33, No. 3, p. vi and vii). The advertisement presents mortality figures compiled from several investigators, some of whom are referenced in the References (21, 22, 42, 83). Serum therapy was considered expensive in its day, an issue addressed in the advertisement by the claim of reduced cost. The dosages recommended in the advertisement are much lower than the 100,000 to 200,000 units per day used in several studies (21, 23). On the basis of the information in the advertisement, the cost of 200,000 units was $120, a considerable expense at the time. These pages were reproduced with the permission of the New York State Medical Society.

and death (78). The injection of serum into the lumbar subarachnoid space produced a therapeutic benefit for the brain but required considerable expertise. The technical difficulties associated with intraspinal administration led to therapy with serum alone administered i.v. (57, 63). Administration by the i.v. route without intraspinal infusion for the treatment of meningitis was controversial at the time (8, 52). The combination of intraspinal and i.v. administration of serum could reduce the rate of mortality from meningococcal meningitis to 5% (95), a rate comparable to that obtained with high doses of penicillin (1). The effectiveness of serum therapy for meningococcal meningitis illustrates that some brain infections can be treated with antibody alone.

Efficacy. Retrospective analysis of the data from several epidemics shows a statistically significant reduction in the rate of mortality for serum-treated patients relative to that for untreated patients (Table 2). When interpreting the data, it is noteworthy that these studies were not case-controlled studies but, rather, were reports of survival of serum-treated patients in comparison with the survival of nontreated patients in the same epidemic. The highly significant P values must be interpreted with caution, given the limitations of the study design. Nevertheless, the data appear sufficiently compelling to conclude that serum therapy markedly reduced the rate of mortality from meningococcal meningitis in those epidemics. On the basis of these results, antimeningococcal serum became standard therapy and was recommended well into the 1940s (24). Between 1920 and 1935 there were several epidemics in which the mortality of serum-treated patients averaged ~50%, leading some to question the value of serum therapy (8, 82, 96). The decreased efficacy of serum therapy in the later epidemics may have resulted from differences in strain virulence and/or antigenic type (8–10, 96). The introduction of sulfonamides in the late 1930s made serum therapy for meningococcal infection obsolete (3).

The exact mechanisms by which antimeningococcal sera mediated therapeutic effects is uncertain. Antimeningococcal sera contained opsonins, agglutinins, and antitoxin antibodies...
Reduced Cost of Type I 
ANTIPNEUMOCOCCIC SERUM Lederle

Improved methods of manufacture permit a reduction in the prices of:

REFINED and CONCENTRATED ANTIPNEUMOCOCCIC SERUM—TYPE I (Lederle)

AS PREPARED BY FELTON

Syringe containing 10,000 units $7.00
Syringe containing 20,000 units $12.00

Packages and Dosage

10,000 Units Type I (One-half size)
20,000 Units Type I (One-half size)

FIRST DOSE — 10,000 units intravenously.
SECOND DOSE — 20,000 units intravenously. Repeat the second dose intravenously at intervals of four to six hours until the temperature falls and beneficial effects are evident.

From the standpoint of lower mortality as well as economy of treatment, Antipneumococcal Serum Type I (Lederle) deserves your consideration.

Literature upon Request


Please mention the JOURNAL when writing to advertisers

FIG. 1—Continued.

(37, 49, 58, 62, 66), and their therapeutic effects were probably due to direct antibacterial effects and enhancement of host immune mechanisms.

NONPHYSIOLOGICAL ANIMAL MODELS IDENTIFIED USEFUL ANTIBODIES

Preclinical testing of antipneumococcal and antimeningococcal sera was done in animal models in which the infection and the antibody were given via nonphysiological routes. Both pneumococci and meningococci enter the human host through the respiratory tree, and their classic clinical presentations are pneumonia and meningitis, respectively. Antipneumococcal sera were developed primarily by using the mouse i.p. model (32, 34, 80, 81, 84), the rabbit dermal pneumonia model (53), and the monkey intratracheal model (19, 23). With regard to the pathogenesis of human infection, none of these animal models of pneumococcal infection can be considered physiologically relevant in humans. In the mouse i.p. and rabbit dermal models, pneumococci are introduced into body compartments not usually exposed to this respiratory pathogen. In the intratracheal model, direct administration of pneumococci into the lower airways circumvents defenses in the nose and upper airways. Similarly, antimeningococcal sera were developed primarily by using the monkey model of intraspinal infection (46-50), which cannot be considered physiological because the bacteria and the serum were introduced directly into the subarachnoid space. Despite little relevance to the pathogenesis of disease in humans, each of these models was effective at identifying potentially useful antibodies for human therapy.

The therapeutic efficacy of passive antibody was difficult to demonstrate in animals with established infections (25, 46, 47, 73). Prophylactic administration of antibody protected mice used in the mouse i.p. model, but the efficacy of therapeutic administration was apparent only in the first 12 h of infection (73). Similarly, it was difficult to demonstrate the efficacy of therapeutic antibody administration against meningococci in guinea pigs and monkeys if serum administration was delayed for more than a few hours (46, 47). However, serum therapy was effective for the treatment of human pneumococcal and meningococcal infections (Tables 1 and 2). The difficulty in demonstrating therapeutic efficacy in these animal models was that the qualities which made them useful for identifying protective antibodies, namely, reliable infection caused by the
inoculation of organisms by nonphysiological routes resulted in fulminant disease with high rates of mortality and had little relevance to human infection. In this regard, pneumococcal infection in mice was uniformly lethal (25), whereas 60 to 70% of patients with pneumonia recovered without therapy (Table 1). Similarly, intraspinal meningococcal infection produced a fulminant meningitis in monkeys (48), whereas 20 to 50% of patients with meningitis recovered without therapy (Table 2). The monkey intratraschale model for pneumococcal pneumonia was more useful for demonstrating the therapeutic efficacies of antibodies, and the beneficial effects of serum could be shown as late as 4 days after infection (19). Presumably, inoculation of pneumococci into the trachea resulted in a less fulminant infection which could be modified by the late administration of antibody. The therapeutic efficacy of antibody could be demonstrated in mice by carefully adjusting the parameters of infection and antibody administration (86).

Morgan and Petrie (76) titrated the size of the inoculum and the dose of serum in a mouse model of i.v. pneumococcal infection and concluded that prophylactic efficacy and curative power were "equally capable of measuring the protective antibody in the serum." Thus, nonphysiological animal models can identify potentially therapeutic antibodies but may be inadequate for demonstrating therapeutic efficacy.

The development of antibody therapy for pneumococcal and meningococcal infections was facilitated by the availability of susceptible animals for the testing of antibody reagents. For pneumococci, the murine i.p. model provided a system in which inocula and antibody dose could be carefully titrated. For meningococci, the monkey intratraschale model (and, later, mucin-treated mice) provided a system in which inoculated animals died of meningococcal infection and in which the effectiveness of antibody reagents could be tested. In these systems the susceptibility of the animal host permitted the use of relatively small inocula, and death resulted from infection and not toxicity related to the inoculum. Animal models in which the routes of infection are nonphysiological may be useful for identifying useful antibodies if the animal host is susceptible (or can be made susceptible) to infection with relatively small inocula.

Animal experiments did not predict the toxicity of serum therapy in humans. The toxicity of serum therapy resulted in anaphylaxis, fevers, chills, and serum sickness (8, 16, 36, 43, 56). Serum sickness was a syndrome characterized by malaise, rash, fever, and arthralgias which usually occurred 7 to 10 days after serum therapy and which was believed to have resulted from immune complexes resulting from host antibody responses to the foreign proteins. Serum sickness was generally a mild self-limited illness which was considered an acceptable side effect (24, 56). Purified antibody preparations were associated with reduced side effects (23, 43). The side effects of serum therapy were the result of immediate and delayed hypersensitivity reactions to animal protein. Recently, the use of mouse MABs in humans has been associated with immunological reactions and the formation of human anti-mouse antibody responses (92). Mouse-human chimeric antibodies are generally well tolerated in humans and are less immunogenic than mouse MABs, but they can elicit human anti-idiotypic responses (69). Newer approaches to the development of fully human MABs may generate less immunogenic antibody molecules.

In summary, passive antibody was an effective therapy for pneumococcal and meningococcal infections. Serum therapy was remarkably successful given the complexity of the immune response and the antigenic variations of bacterial pathogens. The development of serum therapy was characterized by a rapid transition from animal experimentation to human therapeutic trials. The boldness of this strategy was necessitated by an urgent need to develop effective antimicrobial therapies. Clinically useful antibody-based-therapies were identified in animal models in which the routes of infection were nonphysiological. Antibody therapies were effective in humans, despite the difficulty in establishing therapeutic efficacy in animal models. Given the limitations of animal models and the relatively low toxicities of antibodies in therapy (in particular, human antibodies), the experience with the development of serum therapy suggests that once antibody efficacy is established in an animal model, human trials may be justified if the medical problem is particularly urgent. However, the development of serum therapy illustrates the fact that successful translation of antibody therapy from animal models to clinical use required the generation of new knowledge (i.e., the discovery of antigenic variation, need for type specific reagents, etc). Serum therapy in the preantibiotic era provides several
successful precedents for the use of passive antibody therapy against infectious diseases. The efficacies of heterologous polyvalent antitoxin and of the preantibiotic era suggest that passive administration of MAbs, with and without antibiotics, may reduce the rates of mortality from a variety of infectious diseases.

ACKNOWLEDGMENTS

A.C. is supported by a James S. McDonnell Scholar Award and NIH grants AI-33774 and AI-13342. M.D.S. is supported by the Harry Eagle Chair provided by the Women’s Division of the Albert Einstein College of Medicine and by NIH grants CA-39839 and CA-13330-30.

REFERENCES