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Combined enteric and cholera vaccination by the intradermal route

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INTRODUCTION

The value of immunization with a combined typhoid, paratyphoid A and B (TAB) vaccine, in the prevention of enteric fevers, has been accepted for many years by the majority of workers in this field.

Tuft (1931) and (1940) advocated the administration of TAB vaccine by the intradermal route for both primary immunization and for subsequent reinforcement injections. Valentine, Park, Falk & McGuire (1935) reported that appreciably higher agglutinating antibody titres were produced when TAB vaccine was administered by the intradermal rather than by the subcutaneous route. Kamp (1943) noted a marked absence of both general and local reactions in 946 children inoculated intradermally with a combined enteric vaccine and Luippold (1944) reported favourably upon this method of inoculation in volunteer medical students.

Barr, Sayers & Stamm (1959) investigated both the clinical reactions produced, and the protection afforded by a combined enteric vaccine administered to Royal Air Force volunteers either subcutaneously or intradermally. These authors concluded that, whereas the protection afforded by the enteric component was as good when the vaccine was given intradermally as when it was injected subcutaneously, the reactions which occurred with the former technique were significantly reduced, both in number and severity.

As a result of this work, intradermal vaccination for both combined enteric and tetanus (TABT), and enteric (TAB), was introduced at first into the British Army and later into the Royal Navy and the Royal Air Force.

Noble (1963) reviewed all the reactions that occurred during the first 4 years of intradermal inoculation in the British Army, and which were of sufficient severity to warrant the suggestion that the vaccine may have been unduly toxic. He found the incidence of reactions to be negligible.

Ferrán (1885) is accepted, by most modern writers, as the first worker to demonstrate the possibility of an actively induced immunity against cholera. During the 1884 outbreak of cholera in Spain, he was able to demonstrate that guinea-pigs, which survived inoculation with a bile-broth culture of live cholera vibrios, were resistant to the administration of further doses lethal to unprotected animals. Because of impure cultures, Ferrán's vaccination technique gave unsatisfactory results in man and, more often than not, produced excessively severe reactions.

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Camaleia (1888) was the earliest worker to show that protection against cholera could be afforded by the administration of heat-killed suspensions of *Vibrio cholerae*. Further developed by Haffkine (1892), and with only minor refinements since, heat-killed suspensions are to this day the antigen of choice for immunization of non-immune persons against cholera.

Although some workers have from time to time doubted that cholera vaccine provides protection, there seems little doubt that such protection, though of limited duration, is afforded to persons at risk to infection by *V. cholerae* (Pandit, 1948; Mukerjee & Guha Roy, 1962; Archer, 1963) and by the El Tor vibrio (Vella, 1963; Noble, 1964*b* and 1965*a*).

Noble (1964*a*) has reviewed the literature and presented the case for the administration of cholera vaccine by the intradermal route. He concludes that not only is the protection afforded by intradermal inoculation as good as that given by the subcutaneous route, but that a significant reduction in vaccination reactions would occur if the former route were adopted. He goes on to suggest that in order to reduce the number of inoculations required by individuals proceeding overseas, it should be possible to combine intradermal combined enteric prophylactic (TAB) with intradermal cholera vaccine and still retain the freedom from undesirable inoculation reactions, shown separately by the two preparations. This article is a description of laboratory experiments carried out to test this hypothesis.

MATERIALS AND METHODS

Vaccines

Three vaccines were prepared:

(*a*) TAB vaccine, identical with that issued for administration to Service personnel and consisting of *Salmonella typhi* 2000 million, *S. paratyphi* A 1500 million and *S. paratyphi* B 1500 million organisms per ml. suspended in 0·5% phenol-saline.

(*b*) Cholera vaccine (Noble, 1964*a*) consisting of a heat-killed and phenol-preserved suspension of 4000 million *V. cholerae* strain Inaba and 4000 million *V. cholerae* strain Ogawa organisms per ml. in 0·5% phenol-saline.

(*c*) Combined TAB and cholera vaccine (TAB/Ch) containing 5000 million enteric organisms and 8000 million cholera organisms per ml. suspended in 0·5% phenol-saline and obtained by adding equal quantities of double strength TAB and cholera vaccines.

Mice

Because of limitation in supply, mouse strain C/57/Black was used for those experiments in which the challenge organism was *V. cholerae* and mouse strain Ajax albino (Porton) was used for experiments in which an enteric organism was used for challenge purposes. Preliminary experiments suggested that there was little to choose between the mouse strains except that the lethal dose for 100% of animals inoculated (LD 100) in Ajax albino mice was slightly higher (1500 million orgs.) than the LD 100 for C/57/Black mice (1000–1250 million orgs.) when both strains of mice were challenged intraperitoneally with *V. cholerae*.

Active mouse-protection test

This experiment was devised to compare the protection afforded to mice by the administration of each of the three vaccines described above against challenge by *V. cholerae*, *S. typhi*, *S. paratyphi A* and *S. paratyphi B*.

Groups of mice, of average weight 25 g. and with sexes approximately equal in each group, were given two 0·1 ml. intradermal immunizing inoculations of one of the vaccines at an interval of 7 days as described by Noble (1964*a*). Seven days after the second inoculation the mice, together with unprotected mice as controls, were challenged with a predetermined lethal dose of either *V. cholerae* or one of the enteric organisms. Challenge doses were suspended in sterile isotonic saline and administered in a volume of 0·5 ml. injected intraperitoneally. The mice were observed for 72 hr. and survivors noted.

The challenge organisms used in this test and the passive mouse-protection tests which follow were the most virulent of their type available in these laboratories at the time of the experiments. They were: *V. cholerae* Inaba, N.C.T.C. no.7260 (LD 100 approximately 1000×10^6 organisms); *S. typhi*, Ty2 (LD 100 approximately 20×10^6 organisms); *S. paratyphi A*, Kasauli Institute no. B 31/4 (LD 100 approximately 80×10^6 organisms) and *S. paratyphi B*, Millard, recently isolated from a case of paratyphoid meningitis (LD 100 approximately 15×10^6 organisms).

Antiserum

Pooled antiserum was obtained from both rabbits and human volunteers by inoculating them with two intradermal inoculations of 0·1 ml. of the combined enteric and cholera vaccine (TAB/Ch) at an interval of 21 days.

(*a*) Six rabbits (David Bruce Laboratory Lop) were bled to establish a base-line titre of agglutination. No agglutinins for *S. typhi* H, O and Vi or *V. cholerae* O suspensions were detected. Each rabbit was given two intradermal inoculations of 0·1 ml. of the TAB/Ch vaccine, into the skin of the shaved abdominal wall, at an interval of 21 days. The inoculation sites were carefully observed hourly for 12 hr. after injection and thereafter daily, for evidence of untoward reactions. The rabbits were bled 7 days after the second injection. As a check for the effectiveness of the immunizing inoculations agglutination tests (Felix & Bensted, 1954) were performed on each of the six sera separately and upon a Seitz-filtered serum obtained by pooling the six specimens. Agglutinating antibodies to *S. typhi* H, O and Vi and *V. cholerae* O suspensions were detected and their titres expressed as the reciprocal of the highest dilution of serum in the test tube which showed granularity of deposit to the naked eye.

(*b*) Eight human volunteers, from the staff of the David Bruce Laboratories, were found not to have had either enteric or cholera inoculations during the previous 12 months. These volunteers were bled to establish base-line *S. typhi* O and *V. cholerae* O antibody titres. Two 0·1 ml. intradermal inoculations of TAB/Ch vaccine, at an interval of 21 days, were administered to each volunteer into the skin of the outer aspect of the arm behind the posterior border of the distal portion

of the deltoid muscle. The inoculation sites were observed for evidence of undue reactions. Three weeks and three months after the second injection the volunteers were bled again. Serum specimens obtained from the earlier blood specimens were stored in the refrigerator at 4° C. until the later specimens became available. *S. typhi* O and *V. cholerae* O agglutination titres were determined for each serum separately. Finally all the sera were pooled, Seitz filtered and titrated for both *S. typhi* O and *V. cholerae* O agglutinins.

Passive mouse-protection tests

These experiments were devised to demonstrate the protection afforded to mice by either the pooled rabbit or the pooled human antiserum described above against a challenge by a predetermined lethal dose of *V. cholerae* and each of the enteric organisms.

(*a*) Groups of mice, of average weight 25 g. and with the sexes approximately equal in each group, were given one 0·5 ml. subcutaneous injection of pooled rabbit antiserum. Twenty-four hours after the protective inoculation the mice, together with groups receiving serum from unprotected rabbits and unprotected groups as controls, were challenged with a lethal dose of *V. cholerae* (1250×10^6 organisms), *S. typhi* (15×10^6 organisms), *S. paratyphi A* (80×10^6 organisms) and *S. paratyphi B* (15×10^6 organisms). Challenge doses were suspended in sterile isotonic saline and administered in a volume of 0·5 ml. injected intraperitoneally. The mice were observed for 72 hr. and survivors noted.

(*b*) The passive mouse-protection test using human antiserum was performed in exactly the same way as in (*a*) above in which rabbit antiserum was used.

Intradermal skin tests

This group of experiments was designed to study the effects of the combined cholera and enteric vaccine (TAB/Ch) when inoculated into the skin of guinea-pigs, rabbits and human volunteers.

(*a*) TAB/Ch vaccine was inoculated intradermally in two doses of 0·1 ml. at an interval of 21 days. Guinea-pigs and rabbits were injected into the skin of the shaved abdominal wall, and twenty-four human volunteers were inoculated into the skin of the arm, behind the posterior border of the distal portion of the deltoid muscle. The inoculation site and any skin lesions produced were observed and measurements taken at frequent intervals.

(*b*) The effect of repeated intradermal inoculations of TAB/Ch vaccine was studied in both guinea-pigs and rabbits by injecting these animals with doses of 0·1 ml. intradermally at weekly intervals for 9 successive weeks. The injections were all placed into a 1 in. diameter shaved area of abdominal skin but not into the exact site of previous inoculations. The inoculation sites were examined daily for evidence of ulceration, and the presence or absence of indurated nodules assessed by palpation. Daily records of these observations were kept.

Table 1. *Active protection tests in mice inoculated with TAB, cholera or combined TAB/cholera vaccines and challenged with LD* 100 *doses of* Salmonella typhi, S. paratyphi A, S. paratyphi B, *or* V. cholerae

(Active mouse protection test. Average mouse weight 25 g. Sexes approx. equal in each group.)

Mouse strain...	C/57/Black				Ajax albino							
Challenge at 7 days, 0·5 ml. intraperitoneally	*V. cholerae* (Inaba), NCTC 7620 1000×10^6 orgs.				*S. typhi* (Ty 2) 20×10^6 orgs.				*S. paratyphi A* (B 31/4 Kasauli) 80×10^6 orgs.		*S. paratyphi B* (Millard) 15×10^6 orgs.	
Protection 0·1 ml. intradermally × 2 vaccine at 7 days interval	Cholera	TAB	TAB plus cholera	Nil. Unprotected controls	Cholera	TAB	TAB plus cholera	Nil. Unprotected controls	TAB plus cholera	Nil. Unprotected controls	TAB plus cholera	Nil. Unprotected controls
Survivors at 72 hr.	38/40	3/20	37/40	1/20	1/20	39/40	40/40	0/20	40/40	0/20	40/40	0/20

Control virulence tests

Organism...	*V. cholerae* (NCTC 7260)			*S. typhi* (Ty 2)				*S. paratyphi A* (B 31/4)				*S. paratyphi B* (Millard)			
Dose in 0·5 ml. intraperitoneally × 10^6	500	1000	1500	5	10	20	30	40	60	80	100	5	10	15	20
Survivors at 72 hr.	12/20	1/20	0/20	18/20	4/20	0/20	0/20	12/20	3/20	0/20	1/20	12/20	6/20	0/20	0/20
Plate count viability	58 %			62 %				51 %				62 %			

Numerators represent the number of animals to survive. Denominators represent the number of animals inoculated.

RESULTS

Active mouse-protection test

The results obtained in this test are shown in Table 1.

Cholera vaccine and TAB/Ch vaccine, administered intradermally, protected groups of C/57/Black mice equally and well against challenge with live virulent *V. cholerae*, whereas no significant protection was afforded, against a similar challenge, in mice receiving TAB vaccine and in unprotected control mice.

This experiment also showed that groups of Ajax albino mice were almost completely protected against a challenge with virulent *S. typhi* after either TAB vaccine or TAB/Ch vaccine. Those which received cholera vaccine and an unimmunized control group showed no protection against the same challenge.

Finally all the Ajax albino mice in the groups protected with TAB/Ch vaccine survived challenge with either *S. paratyphi A* or *S. paratyphi B* and unvaccinated mice failed to survive a similar challenge.

Antiserum

Agglutination tests performed as a check for the effectiveness of the immunizing inoculation upon the serum of both rabbits and human volunteers gave satisfactory agglutinin titres. The cholera O titres were comparable to the results

Table 2. *Agglutinin titres produced in rabbits after two intradermal inoculations of TAB/Ch vaccine at* 21 *days interval*

	Titres at 7 days			
Rabbit	T 'H'	T 'O'	T 'Vi'	Cholera 'O'
A	640	160	10	320
B	1280	160	5	640
C	640	160	10	1280
D	640	320	10	640
E	320	160	0*	320
F	1280	320	0*	320
Pool A–F and filter	640	160	5	640

Titres expressed as the reciprocal of the highest dilution of serum in the test tube showing granularity of deposit to the naked eye.

* Less than 1:5, the lowest dilution used.

obtained by Noble (1964*a*) when intradermal cholera vaccine was given alone and not in combination. The titres for *S. typhi* antigens were of the same order as those obtained in these laboratories when TAB vaccine was inoculated alone and not in combination with cholera vaccine.

The results obtained for rabbits are shown in Table 2 and for human volunteers in Table 3.

Passive mouse-protection tests

In these experiments most of the mice receiving serum from either TAB/Ch vaccinated rabbits or volunteer humans survived challenge with *V. cholerae* and

Table 3. *Agglutinin responses in human volunteers before, three weeks and three months after the intradermal administration of combined TAB/cholera vaccine*

	S. typhi 'O'			*V. cholerae* 'O'		
Volunteer	Before	3 weeks	3 months	Before	3 weeks	3 months
PF	40	40	320	40	160	320
SO	80	80	160	80	160	80
JH	0*	40	80	80	160	160
CL	20	40	160	40	160	80
TP	20	20	40	0*	20	0*
CM	0*	20	160	0*	20	80
GC	0*	40	320	40	160	320
NH	0*	20	80	20	80	40
Pool and filter		80			160	

Titres expressed as the reciprocal of the highest dilution of serum in the test tube showing granularity of deposit to the naked eye.

* Less than 1:5, the lowest dilution used.

each of the enteric organisms. In contrast, nearly all the mice in the control groups, which received serum from unprotected rabbits and humans or were not given serum, died as a consequence of a similar challenge dose of each of these organisms.

Tables 4 and 5 show the results obtained.

Intradermal skin tests

Combined enteric and cholera vaccine, inoculated intradermally in two doses of 0·1 ml. of a vaccine containing a total of 13,000 million organisms per ml., at an interval of 21 days, failed to produce a reaction of any kind in rabbits and guinea-pigs. The inoculum was absorbed entirely within 24 hr.

In human volunteers similar vaccinations produced local skin responses which were strikingly consistent in appearance, size and duration. At 6 hr. the site of the skin puncture showed a 5–10 mm. firm indurated nodule surrounded by an erythematous flare of a further 5–10 mm. At 12 hr. the central nodule was less well defined, softer to palpation and measured from 25–30 mm. in diameter. The total diameter of the lesion at this time was never greater than 80 mm. From this time onward the reaction gradually resolved, the erythema lessened in intensity and the demarcation between the central and peripheral parts became decreasingly apparent, producing at about 24 hr. an erythematous, soft, brawny, intracutaneous oedema. Resolution rapidly took place, the erythema faded in 48 hr. and by 72 hr. only a small intradermal nodule about 5 mm. in diameter remained. No pyrexia was encountered in any of the twenty-four volunteers. Two subjects complained of slight axillary tenderness but no adenitis was palpable and deep axillary palpation was possible without pain. No volunteer complained of a painful reaction and tenderness to touch was minimal throughout. A series of exercises, including slow and rapid movements, weight lifting and fine movements, such as performing serological agglutinations, showed that in no case was there any loss or impairment of normal function of the vaccinated arm. Several volunteers played

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Table 4. *Passive mouse-protection test using pooled serum from rabbits inoculated with TAB/Ch vaccine by the intradermal route*

(Mice of average weight 25 g. Sexes approx. equal in each group.)

Mouse strain...	C/57/Black			Ajax albino								
Challenge at 24 hr., 0·5 ml. intraperitoneally	*V. cholerae* (Inaba) NCTC 7260 1250×10^6 orgs.			*S. typhi* (Ty 2) 15×10^6 orgs.			*S. paratyphi A* (B 31/4 Kasauli) 80×10^6 orgs.			*S. paratyphi B* (Millard) 15×10^6 orgs.		
Protection 0·5 ml. serum subcutaneously	Pooled immune rabbit serum	Normal rabbit serum	No serum	Pooled immune rabbit serum	Normal rabbit serum	No serum	Pooled immune rabbit serum	Normal rabbit serum	No serum	Pooled immune rabbit serum	Normal rabbit serum	No serum
Survivors at 72 hr.	37/50	8/50	2/50	45/50	9/50	1/50	40/40	4/40	1/40	40/40	5/40	2/40

Numerators represent the number of animals to survive. Denominators represent the number of animals inoculated.

Table 5. *Passive mouse-protection test using pooled serum from human volunteers inoculated with TAB/Ch. vaccine by the intradermal route*

(Mice of average weight 25 g. Sexes approx. equal in each group.)

Mouse strain...	C/57/Black			Ajax albino								
Challenge at 24 hr., 0·5 ml. intraperitoneally	*V. cholerae* (Inaba) NCTC 7260 1000×10^6 orgs.			*S. typhi* (Ty 2) 20×10^6 orgs.			*S. paratyphi A* (B 31/4 Kasauli) 80×10^6 orgs.			*S. paratyphi B* (Millard) 15×10^6 orgs.		
Protection 0·5 ml. serum subcutaneously	Pooled immune human serum	Normal human serum	No serum	Pooled immune human serum	Normal human serum	No serum	Pooled immune human serum	Normal human serum	No serum	Pooled immune human serum	Normal human serum	No serum
Survivors at 72 hr.	22/30	1/20	0/20	28/30	0/20	0/20	20/20	1/20	0/20	20/20	0/20	1/20

Numerators represent the number of animals to survive. Denominators represent the number of animals inoculated.

golf or rugby football 18 hr. after vaccination and when questioned reported no adverse effects either generally or locally at the injection site.

Repeated intradermal inoculation of 0·1 ml. of the 13,000 million organisms per ml., combined cholera and enteric vaccine, at weekly intervals for 9 weeks into a limited area of both rabbit and guinea-pig skins, failed to produce ulceration due to local tissue hypersensitivity. No reactions of any type occurred, except that early in the series it was noted that the inoculum was completely absorbed in approximately 8 hr., whereas after the sixth injection the inoculum tended to require a little longer time, which never exceeded 24 hr., to absorb.

DISCUSSION

Máté, Joó & Pusztai (1964) and Joó *et al.* (1964) have described a combined tetanus toxoid, cholera and typhoid antigen vaccine which showed excellent protection in laboratory tests, including human volunteers, similar to that shown in the experiments described above. These authors inoculated test animals either subcutaneously or intraperitoneally and human volunteers intramuscularly. Although some reduction in reactions was achieved in humans by employing an antigenic extract rather than a bacillary suspension of *S. typhi*, a significant number of both general and local reactions was provoked. Furthermore, at least one drug house in Great Britain is marketing, at this time, a combined enteric and cholera vaccine for subcutaneous or intramuscular administration.

It has been shown that the incidence of severe reactions following the administration of intradermal TAB to Service personnel is negligible (Noble, 1963). An acceptable, painless, local lesion of short duration however occurs (Zuckerman, 1964; Noble, 1965*b*). Noble (1964*a*) has suggested that cholera vaccine administered by the intradermal route is as effective an immunizing agent as when it is given subcutaneously, and reactions are decreased both in numbers and severity when the former route is adopted. The author of a leading article in *Lancet* (1964) appears to support this view.

In the foregoing experiments, it has been shown that in laboratory animals combined TAB and cholera vaccine administered intradermally gives excellent protection against both *V. cholerae* and enteric organisms, while at the same time retaining, in human volunteers, the freedom from severe reactions which would occur if the combined vaccine were administered by the subcutaneous or intramuscular routes.

Many of the reservations held at the time of the adoption of the intradermal technique (Barr *et al.* 1959) have been found to be groundless and vaccination by this method is now a routine procedure in the Services. It is suggested that in view of the increasing number of vaccinations necessary, for example in travellers, following natural disasters and for the rapid mobilization of armies, combined enteric and cholera vaccine (TAB/Ch) by the intradermal route is, where appropriate, preferable to the administration of the two components separately. The intradermal technique rather than either the subcutaneous or intramuscular routes is advocated, not only because the protection afforded appears to be as

good, but also because the incidence and severity of both local and general reactions is considerably less where the former technique is adopted.

Where large numbers are concerned, economy of time and medical manpower become important. Needle-less injectors such as the Microbiological Research Establishment (Porton) 'Hypospray' instrument, already in use for subcutaneous inoculations, have been adapted and are under trial for use with the intradermal technique (Darlow, personal communication). Once the injector has been shown to be satisfactory, the advantages of speed, economy and freedom from reactions obtained by vaccinating, where appropriate, large numbers of non-immune persons with the combined enteric and cholera vaccine by the intradermal route become obvious.

SUMMARY

The protection afforded by a combined enteric and cholera vaccine administered by the intradermal route, and demonstrated by active and passive mouse protection tests and agglutination titres, is excellent and equal to that given individually by the component TAB and cholera vaccines.

In order to reduce the number of injections required and the unpleasant reactions provoked it is suggested that combined enteric and cholera vaccine administered by the intradermal route is of value, where appropriate, for the vaccination of travellers, and for the rapid protection of non-immune persons in the event of natural disasters or mobilization.

REFERENCES

ARCHER, G. T. L. (1963). Miscellaneous immunizations. *Br. J. clin. Pract.* **17**, 690.

BARR, M., SAYERS, M. H. P. & STAMM, W. P. (1959). Intradermal TABT vaccine for immunization against enteric. *Lancet*, i, 816.

CAMALEIA (1888). Quoted by Pollitzer, R. (1959). Cholera. *Wld Hlth Org. Monogr. Ser.* no. 43, p. 294.

FELIX, A. & BENSTED, H. J. (1954). Proposed standard agglutinating sera for typhoid and paratyphoid A and B fevers. *Bull. Wld Hlth Org.* **10**, 919.

FERRÁN, J. (1885). Nota sobre la profiláxis del cólera por medio de inyecciones hipodérmicas de cultivo puro del bacilo virgula. *Siglo méd.* **32**, 480.

HAFFKINE, W. M. (1892). Inoculation de vaccins anticholériques à l'homme. *C. r. Séanc. Soc. Biol.* 9th ser. **4**, 740.

JOÓ, I., PUSZTAI, Z., MÁTÉ, J., RÉTHY, L., ECKHARDT, E. & JUHÁSZ, V. P. (1964). Studies on vaccination against cholera, I. Preparation and laboratory assay of cholera vaccines. *Z. ImmunForsch. exp. Ther.* **127**, 230.

KAMP, M. (1943). Mass typhoid fever immunization—intradermal method. *J. Ind. med. Ass.* **36**, 584.

LANCET (1964). Cholera vaccination. *Lancet*, i, 1147.

LUIPPOLD, G. F. (1944). Typhoid vaccine studies. IX. Intracutaneous versus subcutaneous vaccination for initial immunization. *Am. J. publ. Hlth*, **34**, 1151.

MÁTÉ, J., JOÓ, I. & PUSZTAI, Z. (1964). Studies on vaccination against cholera. II. Vaccination of human volunteers—the reactivity of vaccines. *Z. ImmunForsch. exp. Ther.* **127**, 249.

MUKERJEE, S. & GUHA ROY, U. K. (1962). The vibrios of the recent cholera-like outbreak in Hong Kong. *Br. med. J.* i, 685.

NOBLE, J. E. (1963). Reactions to intradermal TABT and TAB vaccine. *Jl R. Army med. Cps.* **109**, 178.

NOBLE, J. E. (1964*a*). Intradermal cholera vaccination. *J. Hyg., Camb.*, **62**, 11.

NOBLE, J. E. (1964*b*). Vibrio cholerae 'malacca'. *Jl R. Army med. Cps.*, **110**, 15.

NOBLE, J. E. (1965*a*). Trivalent cholera vaccine. *J. Path. Bact.* **89**, 543.
NOBLE, J. E. (1965*b*). Intradermal TAB reactions. *Jl R. Army med. Cps* (in the Press).
PANDIT, C. G. (1948). Composition and efficacy of cholera vaccines. *Proc. 4th Int. Congr. trop. Med. Malar.* **1**, 301.
TUFT, L. (1931). Active immunization against typhoid fever, with particular reference to an intradermal method. *J. Lab. clin. Med.* **16**, 552.
TUFT, L. (1940). Further studies of the intracutaneous method of typhoid vaccination. *Am. J. med. Sci.* **199**, 84.
VALENTINE, E., PARK, W. H., FALK, K. G. & MCGUIRE, G. (1935). A study of agglutinin response to typhoid vaccine. *Am. J. Hyg.* **22**, 44.
VELLA, E. E. (1963). Cholera vaccines and the El Tor vibrio. *Br. med. J.* i, 1203.
ZUCKERMAN, A. J. (1964). Intradermal TAB. *Br. med. J.* ii, 689.