

Title	Protection of Mumps in Children with Various Underlying Diseases : Application of a Live Attenuated Mumps and Trivalent Measles-Rubella- Mumps (MRM) Vaccines in these Children			
Author(s)	Kanesaki, Takumi; Baba, Koichi; Tsuda, Naoki et al.			
Citation	Biken journal : journal of Research Institute for Microbial Diseases. 1986, 29(3–4), p. 63–71			
Version Type	VoR			
URL	https://doi.org/10.18910/82394			
rights				
Note				

The University of Osaka Institutional Knowledge Archive : OUKA

https://ir.library.osaka-u.ac.jp/

The University of Osaka

PROTECTION OF MUMPS IN CHILDREN WITH VARIOUS UNDERLYING DISEASES: APPLICATION OF A LIVE ATTENUATED MUMPS AND TRIVALENT MEASLES-RUBELLA-MUMPS (MRM) VACCINES IN THESE CHILDREN

TAKUMI KANESAKI, KOICHI BABA¹, NAOKI TSUDA, and HYAKUJI YABUUCHI

Department of Pediatrics, School of Medicine, Osaka University, Fukushima, Fukushima-ku, Osaka, 553 Japan

KOICHI YAMANISHI and MICHIAKI TAKAHASHI

Department of Virology, Research Institute for Microbial Diseases, Osaka University, Suita, Osaka, 565 Japan

(Received November 10, 1986)

CUMMARY A live attenuated mumps and trivalent measles-rubella-mumps (MRM) V vaccines have been applied to 887 and 148 children with various underlying diseases at the vaccine clinic of Osaka University Hospital between 1975 and 1985, respectively. Clinical reactions after mumps vaccination occurred in only 7 children (0.8%) and those after MRM vaccination in 28 children (19%), but their underlying diseases were not deteriorated by either vaccination. Clinical follow up study revealed that 2 of the 430 children immunized with mumps vaccine had contracted the disease during 7 year period and 2 of the 123 children immunized with MRM vaccine had contracted clinical mumps or rubella during 3 year period. The seroconversion rates after mumps vaccination were 70% and 61% by the hemagglutination inhibition (HI) test and neutralization (NT) test, respectively, while 94% by the fluorescent antibody to membrane antigen (FAMA) test. Those after MRM vaccination were 87% for measles, 96% for rubella by the HI test and 89% for mumps by the FAMA test. Serological follow up study revealed that antibodies elicited by mumps vaccination were sustained without substantial decline for at least 7 years.

These results suggest that a live attenuated mumps and MRM vaccines are safe and effective in children with various underlying diseases.

INTRODUCTION

Mumps is a common childhood disease. In

healthy children, it is usually mild but may be complicated by meningitis, pancreatitis or orchitis. In children with various underlying

¹ To whom requests for reprints should be addressed.

diseases such as those with acute leukemia, nephrotic syndrome or epilepsy, it may be severe (Henson et al., 1971; Rashid et al., 1977) or deteriorate their underlying diseases (Helin et al., 1983). Therefore it should be considered that prevention of these children against mumps infection is more important than that of healthy children. However, there are few reports that a live mumps vaccine was administered to children with various underlying diseases, except those with acute leukemia reported by Torigoe et al. (1981). We administered a live attenuated mumps and trivalent measles-rubella-mumps (MRM) vaccines to these children in the outpatient clinic of the Department of Pediatrics, Osaka University Hospital between 1975 and 1985. In this paper, we report clinical and serological follow up studies of two vaccines applied in these children.

Furthermore, it was recently reported that seroconversion for mumps occurred in less than two thirds of the recipients of monovalent mumps or MRM vaccine (Sugiura et al., 1982). This reason is considered to be due to not only the insufficient immunogenicity of mumps vaccine but low sensitivity of mumps antibody determination. So, we applied here more sensitive and simplified technique for mumps antibody determination using fluorescent antibody to membrane antigen (FAMA) method.

MATERIALS AND METHODS

1. Study population

Children with various underlying diseases, including neuromuscular, allergic, congenital heart, digestive and renal diseases, congenital malformations and neoplasms have been treated in the outpatient clinic of the Department of Pediatrics, Osaka University Hospital. Children with no history of clinical infection and no antibody against mumps virus were immunized with live mumps vaccine and those with no history of clinical infection and no antibody against each of the three viruses were immunized with MRM vaccine. All the children were under well-controlled conditions at the time of vaccination. Before vaccination, the immune status of individuals was checked by the following tests: skin-tests with PHA, PPD and varicella, assay of antibody activities to these viruses and counts on the number of white blood cells.

2. Vaccines

The live mumps vaccine used was "Biken Urabe Am9", each dose of the vaccine contained approximately 5×10^4 TCID₅₀ of attenuated mumps virus. It was injected singly or in combination with live attenuated measles vaccine "Biken CAM-70" and/or live attenuated rubella vaccine "Biken Matsuura strain".

The MRM vaccine used was trivalent "Biken MRM vaccine (test lot 101)". It contained 2.5×10^5 PFU/ml of CAM-70 measles virus, $10^{3.4}$ PFU/ml of Matsuura rubella virus, $10^{5.7}$ TCID₅₀/ml of Urabe mumps virus. Volumes of 0.5 ml of vaccine were injected subcutaneously into each child.

3. Serologic tests

The antibody response to mumps virus was determined by hemagglutination inhibition (HI) test, neutralization (NT) test and fluorescent antibody to membrane antigen (FAMA) test.

The HI test was carried out as described by Yamanishi et al. (1970). Briefly, after removal of non specific inhibitors by treatment with Kaolin and chick cells, the HI antibody titers of the specimens were determined using 4 units of HA antigen and a 0.5% suspension of chick red cells.

The NT test was performed by cytopathic effect (CPE) method or plaque reduction method. The CPE method was done in microplates using LLC-MK₂ cells as described by Yamanishi et al. (1973). Namely, 0.025 ml of serially two fold diluted serum was mixed with equal volume of antigen containing about 100 TCID₅₀ of mumps virus (Miyake strain) in each well on Corning flat bottom microplates. After incubation for one hour at 37 C, the mixture was kept overnight at 4 C. On the following day 0.1 ml of LLC-MK₂ cells (about 2×10^4 cells/0.1 ml) was added to the serum-antigen mixture. On the 6th or 7th day after incubation, the NT titer was taken as the highest initial dilution of serum which completely inhibited appearance of CPE. The plaque reduction method was done in Vero cells as described by Sakata et al. (1984). Briefly, fourfold serial dilutions of sera were mixed with mumps virus (Enders strain) which was diluted to give 20 to 30 PFU per well. The mixture was kept at 35 C for one hour and inoculated into two wells, each on 24-well (16-mm) Linbro plastic plates. On the 3rd or 4th day after incubation, the NT titer was taken as the serum dilution causing $50^{o}_{.00}$ plaque reduction.

The FAMA test was carried out as described by Baba et al. (1984). Namely, fresh monolayers of Vero cells were infected with mumps virus (Miyake strain) at multiplicity of infection 1 PFU/cell. The cultures were maintained for 48 h until a CPE was observed extensively. Then the monolayers were beaded off into phosphate buffered saline (PBS) and centrifuged at 1,000 g for 5 min. The supernatant was discarded and the pelleted cells were resuspended in medium (a mixture of equal volumes of Eagles' MEM and 199 medium containing 10%heat-inactivated calf serum). Non infected cells, prepared in a similar manner, were used as controls. Aliquots of 0.005 ml of test sera serially diluted in microtiter plates (U plates, Cooke Engineering) were transferred to Terasaki tissue culture plates (60 flat bottom wells, Falcon) and incubated at 37 C for 30 min with an equal volume of mumps virus infected cell suspension (about 8×103 cells) in a humidified atmosphere. The plastic plates were dipped into 180 ml of PBS to remove the supernatant of the cells adhering to the bottom of the plastic. The washed cells were incubated at 37 C for another 30 min with 0.005 ml per well of a dilution of fluorescein-conjugated goat antisera to heavy-chain human IgG (Meloy Chemical Co.). The plates were dipped into PBS to remove the remaining conjugates from the cells, and then the cells were examined in a Nikon inverted fluorescence microscope model TMD. As shown in Fig. 1, fluorescent positive cells for antibody against membrane antigens of cells infected with mumps virus were seen as cells with bright fluorescent



FIGURE 1. Fluorescent positive Vero cells for antibody against mumps virus membrane antigen (×300).

rings around their surface adhering to the plates.

Scropositivity for mumps virus was defined as a titer of $\geq 1:4$ in the NT and FAMA test and a titer of $\geq 1:8$ in the HI test.

The antibody response to measles and rubella viruses were determined by the HI tests, as reported elsewhere (Ueda, 1971; Suzuki et al., 1973) and seropositivity was defined as a titer of $\geq 1:8$.

4. Clinical follow up on vaccinees

Questionnaires were sent to parents of vaccinees. Parents whose children received mumps vaccine were asked whether there had been any cases of mumps in their neighborhood, whether their children had come in contact with cases of mumps, and whether their children had contracted mumps since being vaccinated. Parents whose children received MRM vaccine were asked whether there had been any cases of each of the three diseases in their neighborhood, whether their children had come in contact with cases of each of the three diseases, and whether their children had contracted each of the three diseases since being vaccinated.

If their children had contracted any of the three diseases, we ascertained its clinical course and extent of exposure to the virus by interviewing them when they attended our outpatient clinic.

RESULTS

1. Clinical reactions after vaccination

As shown in Table 1, a total of 887 children classified into 7 groups were immunized with mumps vaccine and a total of 148 children with MRM vaccine. Clinical reactions after mumps vaccination were observed only 7 children (0.8%) 2 to 3 weeks after vaccination. 3 children had mild fever and 4 had slight swelling of the parotid glands. Those after MRM vaccination occurred in 28 children

TABLE 1. Grouping of underlying diseases and Number of children receiving live mumps or MRM vaccine

Grouping of underlying diseases	Number of children receiving live mumps or MRM vaccine		
	mumps vaccine	MRM vaccine	
Diseases of Nervous or Neuromuscular Systems	289	29	
Epilepsy, Febrile convulsion, Hydrocephalus, Cerebral Palsy, Cerebellar			
ataxia, Muscular dystrophy, Myasthenia gravis etc.			
Immunological or Allergic Diseases	152	32	
Asthma, Atopic dermatitis, Egg allergy, Milk allergy, Urticaria, Kawa- saki Diseases, Agammaglobulinemia etc.			
Congenital Heart Diseases	144	17	
Ventricular septal defects, Atrial septal defects, Tetralogy of Fallot,		17	
Single ventricle, Transposition of the great arteries etc.			
Diseases of the Digestive System or Kidney	73	12	
Hepatitis, Intractable diarrhea, Hirschsprung diseases, Congenital		12	
biliary atresia, Lymphoid hyperplasia, Nephrotic syndrome, Glomeru-			
lonephritis etc.			
Diseases of Malignancies	35	13	
Acute leukemia, Malignant lymphoma, Retinoblastoma, Neuroblastoma,			
Wilms tumor, Yolk sac tumor, Hepatoblastoma, Brain tumor etc.			
Diseases of the Endocrine System or Congenital Abnormalities	54	10	
Congenital adrenal hyperplasia, Hypothyroidism, Diabetes Mellitus,			
Addison diseases, Glycogen storage diseases, Mucopolysaccharidosis,			
Down syndrome, 18 trisomy syndrome etc.			
Healthy children	140	35	
Total	887	148	

(19%) with different underlying diseases 5 to 12 days after vaccination. Twenty-four of the 28 children had moderate fever (temperature of \geq 38 C) and 4 had sporadic rash with moderate fever. No other clinical reaction was observed. The incidence of them seemed to be equal in each group. Moreover, in all the children who had them after either vaccination, their underlying diseases were not deteriorated by it.

2. Clinical follow up study after vaccination

Clinical follow up study after mumps vaccination is shown in Table 2. Answers were received to 430 of the 887 questionnaires and divided into 3 groups according to the extent of exposure. There were only 2 cases of mumps during the 7 year period. Among 428 vaccinees who had not contracted mumps, 103 vaccinees had come in contact with cases of mumps (Group I), 70 had encountered mumps epidemics in their neighborhood

TABLE 2. Clinical follow up 1 to 7 years after mumps vaccination

Group		No. of answers	No. of mumps cases
Ι	contact group	104	1
II	epidemic group	71	1
III	no contact and no	255	0
	epidemic group		
total		430	2

(Group II) and 255 had not come in contact with cases of mumps nor had they encountered mumps epidemics in their neighborhood (Group III).

2 cases who contracted clinical mumps after vaccination developed the disease with mild fever and slight swelling of the parotid glands. Their underlying diseases were asthma and congenital heart disease. Blood samples were taken from 2 cases before and after vaccina-

test : A-

test : o---

-

-0

NT (Plaque reduction)

FAMA



FIGURE 2. Serological analysis of two cases who contracted clinical mumps after being vaccinated.

TABLE 3. Clinical follow up 1 to 3 years after MRM vaccination

Disease	No. of contact and/ or epidemic cases cases/answers	No. of contrac- tion cases
Measles	20/123	0
Rubella	23/123	1
Mumps	35/123	1

tion, and after contraction. Antibody titers of these sera were measured by the HI, NT and FAMA test.

As shown in Figure 2, serological analysis revealed that case 1 contracted mumps because he had not acquired immunity after vaccination and that case 2 contracted the disease because he had acquired immunity one month after vaccination but lost it 5 years later.

Clinical follow up study after MRM vaccination is shown in Table 3. Answers were received to 123 of the 148 questionnaires. There were only 2 cases of clinical infection during the 3 year period although the number of contact and/or epidemic cases was small. One child whose underlying disease was asthma contracted clinical rubella with mild fever and generalized eruption when there was epidemics in her neighborhood. Another child whose underlying disease was yolk sac tumor contracted clinical mumps with mild fever and moderate swelling of the parotid glands two weeks after his sister developing the disease.

Blood samples were not taken from 2 cases post vaccination. So, no serological analysis could be done.

3. Antibody response after vaccination

Antibody response after mumps vaccination is shown in Table 4. Blood samples were drawn from 118 of the 428 children who had not contracted clinical mumps after being vaccinated, 4 to 6 weeks after vaccination and measured by the FAMA, HI and NT tests. The seroconversion rate and antibody titer

TABLE 4. Antibody response 4 to 6 weeks after administration of mumps vaccine in 118 children who did not contract the disease after being vaccinated

Method	Seroconversion rate (%)	Antibody titer ^a
FAMA	111/118 (94%)	12.1
HI	83/118 (70%)	6.1
NT^b	72/118 (61%)	3.2

^{*a*} geometric mean titers in all vaccinees.

^b NT test was carried out by CPE method.

TABLE 5. Antibody response 6 weeks afteradministration of MRM vaccine

Virus	Seroconversion rate (%)	Antibody titer ^a
Measles ^b	108/124 (87%)	17.1
Rubella	119/124 (96%)	42.2
$Mumps^c$	101/113 (89%)	11.3

a geometric mean titers in all vaccinees

^b antibody responses to measles and rubella viruses were determined by the HI test.

^c antibody response to mumps virus was determined by the FAMA test.

were highest in the FAMA test. Seven seronegative samples measured by the FAMA test were obtained from the children with agammaglobulinemia and acute leukemia. Therefore, except for these immunologically abnormal children, the seroconversion rate was 100% by the FAMA test.

Antibody response after MRM vaccination is shown in Table 5. Blood samples were taken from 124 of the 148 children 6 weeks after vaccination and measured by the HI and FAMA tests. The seroconversion rates to measles and mumps viruses were approximately 90% and those to rubella virus were 96%.

4. Serological follow up study after mumps vaccination

Serological follow ups were made on the children who had not contracted clinical

Time after vacc	ination	$4 \sim 6$ weeks (N=39)	$1 \sim 2$ years (N=20)	$2 \sim 7$ years (N=14)
I	FAMA	13.0^{b}	11.3	17.1
(contact gr.)	HI	5.7	13.0	8.0
(U)	NT^a	3.2	3.2	3.7
II	FAMA	11.3	9.2	7.5
(epidemic gr.)	HI	6.5	6.1	2.3
	NT	2.8	3.0	3.7
III	FAMA	12.1	11.3	12.1
(no contact and	HI	6.1	6.1	7.0
no epidemic gr.)	NT	3.2	4.0	4.9

TABLE 6. Serological follow up study on the children who did not contract clinical mumps after being vaccinated

^a NT test was carried out by CPE method.

^b each antibody titer was geometric mean titer.

mumps after vaccination. According to the results of clinical follow-ups as shown in Table 2, sequential serum samples taken from these children were divided into 3 groups and measured by the FAMA, HI and NT tests (Table 6). In each group, each antibody titer 4 to 6 weeks after vaccination was not significantly different from that 1 to 2 years after vaccination and that 2 to 7 years after vaccination (p < 0.05). Therefore, it became clear that the FAMA, NT and HI antibody titers were retained without substantial decline for at least 7 years.

Moreover between three groups, there was no significant difference in each antibody titer 4 to 6 weeks, 1 to 2 years and 2 to 7 years after vaccination, respectively (p < 0.05). So, it was considered that some of children of Group III whose parents reported that there had been no contacts and no epidemics had actually come in contact with cases of mumps.

DISCUSSION

The safety and high immunogenicity of live attenuated "Biken mumps vaccine", which was administered to healthy children, were reported by Yamanishi et al. (1970) and Isomura et al. (1973). Clinical and serological follow up studies on these children were also carried out by them (Yamanishi et al., 1971; Isomura et al., 1976), which revealed that protective efficacy of the vaccine was excellent and the HI and NT antibody titers were retained without substantial decrease for at least 4 years.

On the basis of these results, we administered live mumps vaccine to children with various underlying diseases as shown in Table 1, since 1975. A total of 887 children received the vaccine up to 1983. Clinical reactions after vaccination occurred in only 7 children (0.8%) without severe symptoms. This rate was similar to that in healthy children. Moreover, vaccination gave no influence on their underlying diseases. Clinical and serological follow up studies after vaccination, as shown in Table 2 and Table 5, revealed that the protective efficacy of the vaccine was excellent and antibodies elicited by vaccination persisted for at least 7 years without substantial decline in these children.

From these observations, we conclude that a live mumps vaccine was applicable in children with various underlying diseases and gave excellent protective efficacy to these children. The effectiveness and safety of the MRM vaccine have been well documented in foreign countries (Buynak et al., 1969; Smorodintsev et al., 1970; Stokes et al., 1971). In our country, Minekawa et al. (1974) reported the effects of various proportions of measles, rubella and mumps vaccines to determine the quantities in trivalent vaccine required to immunize children effectively. Recently, Sugiura et al. (1982) reported the result of a field trial of the MRM vaccine.

These reports indicated that the observed antibody response and clinical reactions were similar to those following vaccinations with each vaccine alone. So, we applied the MRM vaccine in children with various underlying diseases since 1983 as shown in Table 1. A total of 148 children received the vaccine up to 1985.

Clinical reactions after vaccination were observed 2δ children (19%) without severe reactions. This rate was less than that in healthy children (25.6%) receiving the same lot vaccine as reported by Kakiuchi et al. (1985). Moreover, their underlying diseases were stable after vaccination. The antibody response and protective efficacy following vaccination were excellent, as shown in Table 3 and Table 5. Therefore, we consider that MRM vaccine was useful for prevention of the three diseases in children with various under-

REFERENCES

- Baba, K., Yoshida, M., Tawa, A., Yabuuchi, H., Maeda, K., Takahashi, M. 1984. A simplified immunofluorescence technique for antibody to varicella-zoster membrane antigen (FAMA). Biken J. 27: 23–29.
- Buynak, E. B., R. E. Weibel, J. E. Whitman, Jr., J. Stokes, Jr., and M. R. Hilleman. 1969. Combined live measles, mumps and rubella virus vaccines. JAMA 207: 2259-2262.
- Helin, I., H. Carstensen. 1983. Nephrotic syndrome after mumps virus infection. Am. J. Dis Child. 137: 1126.
- Henson, D., Siegel, S., Strano, A. J., Primack, A.,

lying diseases when applied under careful observations.

Finally, as the protective efficacy against mumps infection after monovalent mumps or MRM vaccination was excellent, the low seroconversion rate for mumps after either vaccination as reported by Sugiura et al. (1982) was considered to be due to the insensitivity of antibody tests to mumps virus. Nowadays enzyme-linked immunosorbent assay (ELISA) is being available as a rapid and sensitive test to determine the antibody response after vaccination (Sakata et al., 1984). However, there remain some problems on the method for antigen preparations and reproducibility of the tests. So, we introduced the FAMA test in this study as a sensitive and simplified test to determine the antibody response after vaccination. As shown in Table 4, the seroconversion rates measured by the HI test and NT test were 70% and 61%, respectively, while those measured by the FAMA test were 94%. As reported previously (Kanesaki et al., 1985), the sensitivity of this test was almost similar to that of ELISA. Moreover, we could simplify the test using Terasaki tissue culture plates and inverted microscope as reported by Baba et al. (1984). Therefore, we consider that the FAMA test is one of the useful methods to determine the antibody response after mumps vaccination.

Fuccillo, D. A. 1971. Mumps virus sialoadenitis, An autopsy report. Arch. Pathol. 92: 469-474.

- Isomura, S., Asano, Y., An, S., Miyata, T., Suzuki, S. 1973. Studies on live attenuated mumps vaccine. I. Comparative field trials with two different live vaccines. Biken J. 16: 39-42.
- Isomura, S., Ozaki, T., Morishima, T., Nagayoshi, S., An, S., Suzuki, S. 1976. Studies on live attenuated mumps vaccine. II. Follow-up study on the efficacy of Biken vaccine. Biken J. 19: 115–118.
- Kakiuchi, T., Nambu, M., Inoue, Y., Hayakawa, Y., Yamanishi, K., Ueda, S., Takahashi, M.,

70 BIKEN JOURNAL Vol. 29 No. 3-4 1986

Okuno, Y. 1985. Clinical and serological evaluation of a trivalent measles-rubella-mumps (MRM) vaccine. Jpn. J. Pediatrics 38: 1189–1193. [In Japanese]

- Kanesaki, T., Baba, K., Tsuda, N., Yabuuchi, H. 1985. Determination of mumps antibody by fluorescent antibody to membrane antigen (FAMA) test. Clin. Virol. 13: 242–246. [In Japanese]
- Minekawa, Y., Ueda, S., Yamanishi, K., Ogino, T., Takahashi, M., Okuno, Y. 1974. Studies on live rubella vaccine. V. Quantitative aspects of interference between rubella, measles and mumps viruses in their trivalent vaccine. Biken J. 17: 161–167.
- Rashid, A., Cress, C. 1977. Mumps uveitis complicating the course of acute leukemia. J. Pediatr. Ophthalmol. 14: 100–102.
- Sakata, H., Hishiyama, M., Sugiura, A. 1984. Enzyme-linked immunosorbent assay compared with neutralization tests for evaluation of live mumps vaccines. J. Clin. Microbiol. 19: 21-25.
- Smorodintsev, A. A., Nashibov, M. N., Jakovleva, N. V. 1970. Experience with live rubella virus vaccine combined with live vaccines against measles and mumps. Bull. WHO 42: 283–289.
- Stokes, J., Jr., Weibel, R. E., Villarejos, V. M., Arguedas, J. A., Buynak, E. B., Hilleman, M. R. 1971. Trivalent combined measles-mumps-rubella vaccine. Findings in clinical-laboratory studies. JAMA 218: 57–61.
- Sugiura, A., Ohtawara, M., Hayami, M., Hishiyama, M., Shishido, A., Kawana, R., Hirayama, M., Makino, S., Kimura, M., Isomura, S., Takahashi,

M., Kawakami, K., Matsuyama, S. 1982. Field trial of trivalent measles-rubella-mumps vaccine in Japan. J. Infect. Dis. 146: 709.

- Suzuki, N., Minekawa, Y., Osame, J., Ueda, S., Ogino, T., Yamanishi, K., Takahashi, M., Okuno, Y., Kan, T., Onishi, N., Koyama, K. 1973. Studies on live rubella vaccine. III. Field trials with vaccines attenuated by serial passages in primary pig kidney cells. Biken J. 16: 149–154.
- Torigoe, S., Hirai, S., Oitani, K., Ito, M., Ihara, T., Iwasa, T., Kamiya, H., Sakurai, M. 1981. Application of live attenuated measles and mumps vaccines in children with acute leukemia. Biken J. 24: 147–151.
- Ueda, S. 1971. Comparison of measles antibody titers measured by the micro- and macro-methods. Biken J. 14: 155-160.
- Yamanishi, K., Takahasbi, M., Kurimura, T., Ueda, S., Minekawa, Y., Ogino, T., Suzuki, N., Baba, K., Okuno, Y. 1970. Studies on live mumps virus vaccine. III. Evaluation of newly developed live mumps virus vaccine. Biken J. 13: 157-161.
- Yamanishi, K., Takahashi, M., Kurimura, T., Ueda, S., Suzuki, N., Baba, K., Okuno, Y. 1971. Studies on live mumps virus vaccine. IV. Clinical and serological follow-up 1 year and 3 years after vaccination. Biken J. 14: 259–265.
- Yamanishi, K., Takahashi, M., Ueda, S., Minekawa, Y., Ogino, T., Suzuki, N., Okuno, Y. 1973. Studies on live mumps virus vaccine. V. Development of a new mumps vaccine "AM9" by plaque cloning. Biken J. 16: 161–166.