This interim report on prophylaxis of rabies in man reviews briefly the development of experimental studies with laboratory animals and presents results to date of tests of such experimental methods applied to man.

Rabies Prophylaxis in Man

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IN GENERAL, research in rabies prophylaxis has developed along two lines. One has been aimed at improving the effectiveness of rabies prophylaxis in man; the other has been concerned with methods of reducing the severity of reactions to vaccine prophylaxis, especially reactions of the dangerous neurological type.

Efforts to improve the efficacy of prophylaxis were stimulated by the demonstrated failure of vaccine in preventing rabies after severe exposure. Experience in Middle East countries, such as Iran, showed that persons severely bitten by rabid wolves developed rabies at the same high rate (40 percent) whether given a full course of potent vaccine or no treatment (*1*). Research in recent years has centered on the use of rabies antiserum as an adjunct to vaccine. In laboratory experiments with modern quantitative techniques, antiserum alone gave better results against experimental infection than vaccine alone; furthermore, antiserum alone tended to prolong markedly the incubation period. Best protection was obtained when antiserum was used together with a course of vaccine (*2*, *3*).

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In order to determine whether doses of vaccine and antiserum practical for use in man would give similar results, the World Health Organization's Expert Committee on Rabies set up two studies. The first study observed effects on normal human volunteers given various schedules of vaccine either exclusively or combined with a single dose of antiserum. Evaluation of efficacy was based on the serum neutralizing antibody titers developing in the blood of the volunteers at various intervals during and after the course of immunization. A summary of some of the results of this experiment, the details of which have been published by WHO (*4*), is given in figures 1 and 2. Ten persons were observed in each treatment group, and the points on the graph represent the average serum titers of the 10 patients at the specified number of days from the start of treatment.

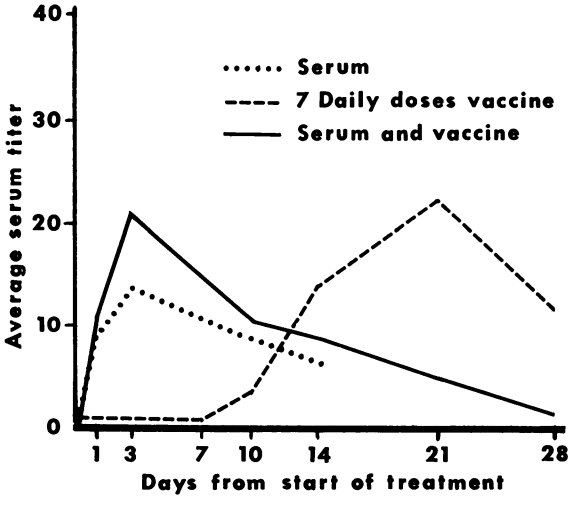
In figure 1 it can be seen that the individuals receiving serum alone in the form of a single injection of antirabies rabbit serum had demonstrable antibody 1 day later; the antibody gradually decreased in titer to the 14th day. The group receiving 7 daily doses of vaccine showed first evidence of antibody response at 10 days, but significant levels did not appear until the 14th day and then persisted. However, if antiserum was given 1 day before the start of vaccine, the pattern of antibody level was similar to that in the group which

received serum alone, and there was no evidence of an active antibody response to the vaccine after the 14th day.

Figure 2 shows comparative results when 12 daily doses of vaccine were given. Again, active antibody production due to vaccine alone was not apparent until the 14th day. It was quantitatively higher than after only 7 doses, and it persisted throughout the 28-day period. Here the combination of that same schedule of vaccine with a single dose of serum gave a significant antibody level early because of the passive immunization with antiserum and late because of active immunization with the vaccine.

These experiments and others indicate that there is some interference with the antigenic effect of relatively low doses of vaccine when serum antibody is given early, but this inhibition can be overcome if more vaccine is given over a longer period of time. On the other hand there was no evidence that the vaccination neutralized the antibody in the passively administered antiserum. Obviously, figure 2 shows the type of response in which we are interested. We would like to have antibody present throughout the course of treatment: antibody introduced early by the antiserum that is given passively; antibody produced late by the active response of the individual to the vaccine.

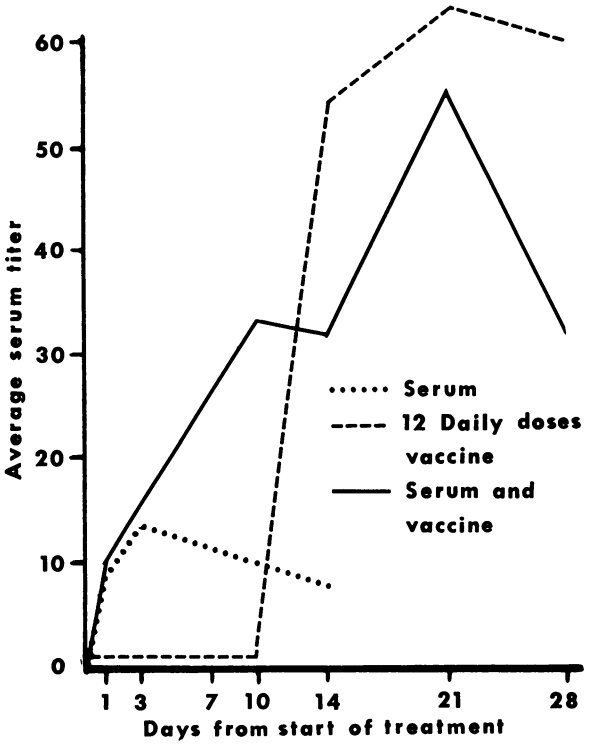
Figure 1. Comparison of neutralizing antibody response of human volunteers to rabies antiserum alone, 7 daily doses of vaccine, and combined treatment.



SOURCE: Reference 4.

NOTE: Each point on the chart represents the average antibody levels of 10 persons and is the reciprocal of the serum dilution which neutralizes 32 LD_{50} of virus.

Figure 2. Comparison of neutralizing antibody response of human volunteers to rabies antiserum alone, 12 daily vaccine doses, and combined treatment.



SOURCE: Reference 4.

NOTE: Each point on the chart represents the average antibody levels of 10 persons and is the reciprocal of the serum dilution which neutralizes 32 LD_{50} of virus.

Field Test in Iran

The second study set up by the WHO Expert Committee on Rabies for the use of antiserum in man was an actual field test in exposed individuals. Because of past experience of vaccine failures in prophylaxis following bites by rabid wolves in Iran, test arrangements were made with Dr. M. Baltazard at the Pasteur Institute in Teheran. Groups of individuals subsequently exposed were to be divided into treatment groups so that comparable exposures could be treated either with vaccine alone or

with vaccine plus antiserum. As frequently happens no such group of exposed individuals became available for some time. When they did more than 3 years later, the local physician was not aware that the bites represented exposure to rabies since the wolf was not apprehended. No treatment was administered until the 20th day when the first of the 17 bitten individuals developed rabies. Although vaccine treatment of the rest of the group was started at this time, the interval between exposure and treatment was so long that the entire group can be considered as receiving no specific prophylaxis. Forty percent of these individuals died of rabies.

One year later an ideal group presented itself, and the story of the exposure is indeed a dramatic one (*5*). In August 1954, several busloads of people stopped to spend the night at a small town in the forests some 500 kilometers from Teheran. Because of the heat many bus passengers and townspeople slept outdoors on the ground or on open porches and balconies. At midnight a scream was heard from an orchard on the edge of town because a large wolf had attacked one of the sleeping men. Within a very short time the wolf bit several others in the orchard, circled a bit and entered the center of the village, where he attacked and bit several people on the street. He even entered some homes slashing people asleep on a balcony and proceeded across town attacking more along the main road. The wolf finally bit six cows in a nearby field and was killed as he lunged at a horse and rider. A total of 29 people, 1 dog, and 6 cows had been bitten during a 5-hour period.

All exposed individuals plus the dead wolf were placed in a commandeered truck and taken to the Pasteur Institute in Teheran, where treatment was started within 30 hours after exposure. The wolf was proved rabid by isolation of rabies virus from his brain. His saliva apparently had been infectious throughout the period of his attack since the first person bitten died of rabies as did two of the cows which were bitten just before the wolf was killed. The patients were divided into treatment groups (table 1). Only the 18 bitten on the head or neck are important in evaluating the treatment results since past experience in Iran had indicated that most rabies deaths occurred after this type of exposure.

A group of 5 patients, 4 of whom had severe bites, were treated with two inoculations of serum given 4 days apart plus a full 21-day course of vaccine; no cases occurred. Another group of 7 patients received one dose of serum on the first day, followed by 21 doses of vaccine; 1 out of the 7 died from rabies. The third group, consisting of 5 individuals, 4 of whom had been severely bitten on the head, received the course of vaccine alone. Of this group, 3 died of rabies. One boy, patient No. 27 listed separately in table 1, was bitten directly through his skull and received practically an intracerebral inoculation of saliva from the rabid wolf. He was unconscious on arrival

Table 1. Mortality of patients bitten by rabid wolf, Iran, 1954

Group	Number of patients	Number of patients with severe exposure	Location of bites	Treatment	Mortality
A	5	4	Head	2 inoculations of serum plus vaccine.	0/5
B and patient No. 28.	7	7	Head	1 inoculation of serum plus vaccine.	1/7
C	5	4	Head	Vaccine only.	3/5
Patient No. 27	1	1	Meninges	6 inoculations of serum plus vaccine.	0/1
D	4	0	Trunk and extremities	1 inoculation of serum plus vaccine.	0/4
E and patient No. 29.	7	0	Trunk and extremities	Vaccine only.	0/7

SOURCE: Reference 5.

Table 2. Neutralizing antibody levels in serum samples of group A patients exposed to head and neck bites of rabid wolf and treated with two doses of serum and complete course of vaccine

Patient No.	Outcome	Antibody titers on days after start of treatment [1]															
		1	3	4	5	6	7	8	10	13	15	19	21	25	29	33	41	53
A1	S	8	+	---	22	---	+	---	22	---	22	---	22	---	5	---	+	12
A2	S	13	+	---	32	---	+	---	22	---	22	---	12	---	6	---	+	3
A3	S	tr	---	---	13	---	+	---	66	---	30	---	76	---	191	---	+	76
A4	S	tr	+	---	---	32	+	---	22	---	22	---	12	---	6	---	---	tr
A5	S	6	+	---	112	---	+	---	10	---	17	---	22	---	22	---	+	8
27 [2]	S	---	---	19	---	38	---	99	112	99	+	30	---	32	---	22	---	---

[1] The figures shown are the reciprocals of the serum dilutions, representing the 50 percent end point in the neutralization test against 8–46 LD_{50} of virus.

[2] Patient 27 received 6 injections of serum and a complete course of vaccine.

S=Survived. D=Died of rabies.
+=Virus neutralized by undiluted serum; no titration of antibodies done.
tr=Partial neutralization of virus at a dilution of less than 1:5 of serum.

SOURCE: Reference 6.

at the Pasteur Institute. Because of his severe exposure, he was given not only the course of vaccine but also a dose of serum on alternate days for 6 doses. That boy also survived. The other cases listed at the bottom of table 1 are not significant because they were bitten elsewhere than on the head, and we know that even without treatment most of them probably would have survived.

Dr. Hilary Koprowski and I received the serums from all these patients (blood samples were taken at intervals for a period of 50 days by the group in Teheran), and quantitative neutralization tests for rabies antibodies were run on certain of the samples (*6*). Group A, which received 2 doses of serum plus a course of vaccine, showed no antibody, of course, before treatment. Antibody was demonstrated on the first day and persisted at good levels on the 5th and the 10th days. In serum specimens collected at later periods when antibody is not expected to persist, we found that these individuals were developing active antibody as a result of the vaccine. All of these people survived; all of them had antibody (table 2).

In group B, which received only one dose of serum plus a course of vaccine, the one person who died of rabies showed no active antibody response to the vaccine although he had shown antibody levels early after the inoculation of antiserum (table 3).

Of the 5 patients in the control group, which received vaccine only, 3 died of rabies (table 4). There seems to be very little or no correlation between the amount of antibody produced by the active immunization and the subsequent outcome of the disease. One patient who died had no antibody at any time, yet another with fatal outcome had an excellent antibody response. Of course, we have to remember that the antigen of the street virus introduced by the rabid wolf multiplies during the incubation period and can also call forth an antibody response. So it is very difficult in such a study of exposed individuals to evaluate the antibody levels appearing late in the course of immunization. Also in this vaccine group was an individual who survived and yet at no time during the entire course of treatment had any antibody. In my opinion, this individual is one of those persons who do not respond to antigens and I think, undoubtedly, never had an effective exposure to rabies.

As a result of the earlier experimental data and these field trial results, the WHO committee has recommended the routine use of a single dose of antiserum followed by a course of at least 14 doses of vaccine for all severe exposures (*7*). Two further practical points on the use of antiserum should be mentioned. Experimental results as yet untested in the field indicate that local infiltration of part of the total dose of antiserum about the bite wound increases its effectiveness. Finally, the anti-

Table 3. Neutralizing antibody levels in serum samples of group B patients exposed to head and neck bites of rabid wolf and treated with one dose of serum and complete course of vaccine

Patient No.	Outcome	Antibody titers on days after start of treatment [1]												
		1	3	5	7	10	12	15	17	19	21	29	41	53
B1	S	6	+	22	+	6	------	tr	------	------	8	tr	tr	13
B2	D	tr	+	20	+	tr	tr	tr	tr	tr	------	------	------	------
B3	S	6	+	10	+	6	------	5	------	------	5	13	6	------
B4	S	tr	+	8	+	13	------	18	------	------	6	8	+	13
B5	S	0	+	15	------	13	------	tr	------	------	5	tr	------	------
B6	S	tr	tr	6	tr	8	------	8	------	------	30	67	------	112

[1] See footnote 1 and legend, table 2.

SOURCE: Reference 6.

Table 4. Neutralizing antibody levels in serum samples of group C patients exposed to head and neck bites of rabid wolf and treated with complete course of vaccine only

Patient No.	Outcome	Antibody titers on days after start of treatment [1]													
		1	3	5	7	10	15	19	21	25	29	33	41	45	53
C1	D	0	0	0	0	0	0	22	50	85	66	-----	-----	-----	-----
C2	S	0	0	0	0	0	0	tr	5	13	6	-----	19	-----	5
C3	D	0	0	0	0	0	0	-----	0	0	-----	-----	-----	-----	-----
C4	D	0	0	0	0	0	0	tr	-----	18	27	22	15	89	18
C5	S	0	0	0	0	0	0	-----	-----	-----	0	-----	0	-----	0

[1] See footnote 1 and legend, table 2.

SOURCE: Reference 6.

serum presently available commercially in this country is a concentrated horse serum, and tests for sensitivity must be performed before its use. Serum sickness in from 5 to 20 percent of the treated cases is to be expected.

Vaccination Schedules

Research effort in recent years aimed at reducing the severity of prophylactic measures and reactions to them stems from two facts.

First, the currently used rabies vaccine is still a crude biological product. The vaccine is a heavy suspension of rabies infected rabbit brain in which the virus has been inactivated by various chemical or physical agents. Fourteen to twenty-one or more injections are made with this crude brain emulsion in as many days. All treated patients get local reactions, usually of moderate severity. Also, the necessity of a large number of daily visits to the physician requires actual physical residence at a center where treatment is available. This is a hardship for those patients whose homes are far from a medical facility.

The second fact concerns severe and dangerous reactions to the vaccine, the most important of which involve the central nervous system. Experimental work by Morgan (*8*), Rivers and associates (*9*), and others strongly suggests that these reactions are directly related to the multiple injection of the brain tissue contained in rabies vaccine. The relative importance of this postvaccinal problem of encephalitis or paralysis varies in different parts of the world. In general, we in the United States are more concerned than rabiologists in other countries. Other than concern about the influence of poor reporting on the evaluation of the severity and frequency of reactions, the chief reason for American interest probably lies in the fact that, with our low mortality from rabies, it is estimated we have more cases of central nervous system reactions to vaccine than deaths from

the disease. Ten to twenty-five percent of these reactions are fatal.

Because of the severity of the procedure, the practical difficulties involved, and the fact that the occurrence of postvaccinal CNS reactions seem related to the number of injections of brain tissue, investigations are under way to determine the feasibility of reducing the number and size of vaccine injections. Fox and associates (*10*) have shown that 3 doses given 5 days apart give good serum antibody levels in man. This type of study in nonexposed human volunteers is currently being investigated further by the WHO committee. Tables 5 and 6 show the results of our experiments in mice in testing the relative efficacy of different schedules of immunization (*11*). Table 5 shows the effect of variations in number and spacing of vaccine doses when the total dose is the same for all groups, and table 6 evaluates the same effect when each individual dose is the same in all groups. In either case, it is obvious that daily doses did not give much better protection against intracerebral virus challenge or produce antibodies at a significantly higher level than some of the less severe schedules. In general, these results indicate the importance of 2 or 3 early primary doses followed by a booster dose at or after the 10th day.

Table 5. Effect of same total vaccine dose given in various schedules on immunity to intracerebral challenge and neutralizing antibody response in mice [1]

Group	Dose (ml.)	Days	LD_{50} protection (logs)	Serum dilution 50 percent neutralization
A	0.1	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12.	3.7	187
B	.4	1, 5, 10	3.4	56
C	.6	1, 10	2.9	80
D	.2	1, 3, 5, 8, 10, 12.	4.1	68
E	.6	1, 8	2.2	32
F	.4	1, 2, 8	2.2	46
G	.3	1, 2, 3, 8	2.2	56
H	.3	1, 2, 3, 10	3.3	187
K	.4	1, 2, 10	2.6	95
L	.3	1, 2, 8, 12	3.3	279
M	.3	1, 2, 3, 12	3.3	279
O	.4	1, 2, 12	2.6	162

[1] Ultraviolet irradiated vaccine was used diluted to 0.6 percent suspension. Intraperitoneally immunized mice were bled on the 14th day and challenged intracerebrally on the 15th day.

SOURCE: Reference 11.

Table 6. Effect of various vaccine doses and schedules on immunity to intracerebral challenge and neutralizing antibody response in mice [1]

Group	Days	LD_{50} protection (logs)	Serum dilution 50 percent neutralization
A	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12.	4.2	944
B	1, 5, 10	3.3	1,084
C	1, 3, 5, 8, 10, 12	>4.7	>3,125
D	1, 2, 3, 10	3.2	625
E	1, 2, 8, 12	2.1	64
F	1, 2, 3, 12	3.5	71
G	1, 10	1.1	90
H	1, 2, 12	1.4	43
K	1, 2, 10	2.3	215

[1] Phenolized vaccine was used diluted to 1 percent suspension. Intraperitoneally immunized mice were bled on the 14th day and challenged intracerebrally on the 15th day. All individuals were given 0.2 ml. of vaccine.

Avian Embryo Vaccines

Other studies have attacked the problem of reactions to brain tissue vaccines from the standpoint of finding effective vaccines in which the factor responsible for these reactions has been removed or of making vaccines from material other than brain tissue. The former has been accomplished in the laboratory but has not proved practical in vaccine production (*2*). A fair amount of research has been carried out by Fox (*10*), by the WHO rabies committee (*4*), and by Dr. Hilary Koprowski on the possible use in man of live attenuated chick embryo rabies vaccine similar to that currently used in canine immunization. Thus far, this vaccine has been given to several hundred individuals with no deleterious effect. However, in contrast to results in dogs a single dose does not produce immunity in man as indicated by a serum antibody response. Several booster doses are required before antibody becomes demonstrable. In other words, in the dog this vaccine acts as a live virus vaccine with multiplication of the attenuated virus, but in man it acts as an inactivated virus vaccine, and the

virus probably does not multiply. Fox has shown good antibody response after 3 intradermal doses given 5 days apart, and Koprowski has evidence that a single intradermal dose given to individuals who have received rabies vaccine even several years previously will have a booster effect and result in a prompt antibody rise.

In a recently developed vaccine, now commercially available, the rabies virus is produced in duck embryos and is inactivated by beta propriolactone (*12*). This "killed" virus vaccine when used in multiple doses comparable to the brain tissue vaccines is effective in protecting experimental animals against virus challenge and in producing an antibody response in man.

In conclusion, there is hope in the future for the development of a rabies vaccine devoid of the potentiality of producing severe or fatal central nervous system reactions, but by far the most significant recent accomplishment in the field of rabies prophylaxis in man has been the further development of rabies antiserum and the demonstration in a clinical trial of its superior efficacy when combined with vaccine.

Summary

Previous experimental evidence of the greater efficacy of rabies antiserum as an adjunct to rabies vaccine over that of vaccine alone has been confirmed in a dramatic clinical trial. Two doses of antiserum given 4 days apart together with 21 daily doses of vaccine completely protected 5 individuals severely exposed by the bite of a rabid wolf in Iran. One of seven comparably exposed patients who received one dose of antiserum plus a course of vaccine died of rabies, while 3 of 5 receiving vaccine alone succumbed. Serum antibody studies of blood samples of these individuals showed the presence of antibody early and late in the course of treatment in those receiving both serum and vaccine but only in the late period when vaccine alone was used.

Experimental investigations in animals and in man suggest that the number of doses of vaccine may be reduced with proper spacing without markedly reducing its effectiveness. The importance of a booster dose given 10 days after the primary doses was apparent.

Attempts to eliminate severe neurological reactions to rabies vaccine have stimulated research with two types of vaccine produced from avian embryos. In laboratory tests, both appear comparable to the currently used brain tissue vaccine.

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Taking Care of Diabetes

ELEVEN FILMSTRIPS IN SOUND AND COLOR

Audience: The patient and his family.

Availability: Loan—Communicable Disease Center, Public Health Service, 50 7th Street, NE., Atlanta, Ga. Purchase—United World Films, Inc., 1445 Park Avenue, New York 29, N. Y.

Designed to help the diabetic patient and his family, each of the 11 filmstrips depicts some phase of the problem of diabetes control. These 35-mm. filmstrips, originally produced in 1950, were reissued in 1956.

What Is Diabetes? *47 frames, 8½ minutes.* Helps a patient develop a wholesome attitude toward diabetes by giving him a better understanding of his condition and how it can be controlled; emphasizes the need of cooperation with the doctor, acceptance of the major responsibility for management and control of his diabetes, and maintenance of a positive point of view.

Eating for Good Health. *46 frames, 7 minutes.* Stresses the role of food in controlling diabetes and maintaining good health; aids the diabetic and his family in gaining a better understanding of the patient's condition and the foods he may eat.

Insulin and Its Use. *69 frames, 13 minutes.* Attempts to give the patient an understanding of what insulin is, where it comes from, how it functions in the body, and why some people with diabetes need to take it; shows the different kinds of bottle insulin; and illustrates a method of care and handling of the equipment necessary for the injection of insulin, and demonstrates the technique for injecting insulin.

Planning Good Meals. *52 frames, 9 minutes.* Shows the patient how he can plan a wide variety of meals by using his meal plan and exchange list and explains the function of the various kinds of foods—carbohydrates, proteins, fats, vitamins, and minerals—in maintaining health.

Buying Good Food. *50 frames, 8 minutes.* Explains how certain foods are arranged in groups called exchange lists, illustrates how the lists help patients in buying food in ordinary grocery stores, and shows the variety of foods to be selected and their relative value.

Insulin Reaction. *39 frames, 6 minutes.* Helps the person with diabetes recognize and understand the symptoms of an insulin reaction; shows some of the causes, ways of preventing, and effective means of emergency treatment; and points out the serious effects reactions can have on a patient.

Tests in Diabetes. *38 frames, 6½ minutes.* Shows the relation of urine sugar testing to the degree of control of diabetes, how the patient can test his urine for sugar, and outlines action the patient can take when tests are repeatedly positive.

Cooking Good Meals. *40 frames, 8 minutes.* Gives the recommended practices that apply to cooking for the whole family, shows they apply also to the diabetic, and notes different ways of preparing food.

Diabetic Coma. *33 frames, 5 minutes.* Gives some understanding as to what diabetic coma is and how it develops; discusses its seriousness, the need of seeing a doctor when the danger signs appear, and the importance of daily urine and sugar tests, following a meal plan, and taking just the right amount of insulin in order to avoid diabetic coma.

Care of the Feet. *49 frames, 8½ minutes.* Demonstrates why the diabetic patient should take proper care of his feet, how to select proper fitting shoes and socks, and what exercises aid in maintaining good blood circulation and health of the feet; how other serious difficulties from poor circulation, injuries, and infections can be prevented.

Selecting Meals for All Occasions. *52 frames, 8 minutes.* Emphasizes the use of certain basic foods, such as meat, fruit, and milk, for providing variety in food selection and assuring the selection of the right foods to eat in different situations—when one is ill, going on a picnic, eating at a friend's house or at a restaurant, or taking lunch to work.