nase and ornithine transcarbamylase, showed no correlation with the growth rate or histological type. From a survey of isozyme patterns of several enzymes he tried to quantitate malignancy of these experimental hepatomas; he studied several isozymes including lactate dehydrogenase, glucose-6-phosphate dehydrogenase, glutamic-oxalacetic transaminase, glutamic-pyruvic transaminase, malate dehydrogenase, and α-glyceroaldehyde dehydrogenase. Ono showed that the isozyme pattern of the LDH (lactic dehydrogenase) of all ascites hepatomas tested retained essentially the normal pattern of rat liver, and that the variation among the 53 Yoshida strains was caused in part to a gradation of differentiation. The "minimal-deviation" type hepatomas exhibited the well differentiated pattern similar to that of normal adult liver. Although one minor peak of the glucose-6-phosphate dehydrogenase isozyme was deleted in a few cases no abnormal isozyme pattern was observed. Ono proposed three biochemical parameters which could possibly be used to quantitate malignancy of experimental hepatomas: (i) enzyme activities which correlated either positively or negatively to the growth rate (glucose-6-phosphatase, and glutamate dehydrogenase), (ii) independence of inducible or suppressible enzymes from host control (tryptophan pyrrolase and glucose-6-phosphate dehydrogenase), and (iii) systemic effect of hepatomas on the host liver enzymes, such as catalase activity.

In his closing remarks W. Nakahara (National Cancer Center Research Institute, Tokyo) referred to the historical background of his early studies on the catalase activity in tumor-bearing animals. He emphasized that extensive studies on the cell membrane might be desirable for better understanding of malignancy, because less adhesiveness and invasiveness were common during the oncogenic processes.

The symposium clearly established that a continuous spectrum of hepatomas revealing different growth rates represented many degrees of progression of cancer cells and that more studies were vital. The clinical symptoms or signs, growth rate, transplantability, and others, used in the past must now include both the biochemical and biological approaches. These newer approaches have now opened up new frontiers of cancer research. The etiology of cancer, however, is still far beyond our present knowledge. Rather, all findings presented so far appear to be reflected from or due to adaption to the rapid division of cancer cells. No evidence was presented indicating that the primary event of oncogenic processes is from somatic mutation. It is still unknown whether or not all alterations so far observed in cancer cells are due to a common but single alteration in the enzyme-synthesizing systems or to multiple combinations of alterations in the regulatory mechanism for individual enzyme-synthesizing systems. Disturbance or unbalance might be present at higher levels of regulatory mechanisms of cellular activities, such as differentiation, development, or growth rather than a single metabolic step.

The proceedings of this symposium will be published shortly in the Japanese journal of cancer research, *GANN*, edited by the Japanese Cancer Society.

YASUTOMI NISHIZUKA
OSAMU HAYAISHI
Department of Medical Chemistry, Kyoto University, Kyoto, Japan
HAROLD P. MORRIS
National Cancer Institute, National Institutes of Health, Bethesda, Maryland

Swine in Biomedical Research

In recent years swine have been used increasingly in biomedical research. To explore the basis for and extent of this use, as well as to provide a firm basis for the future use of swine in biomedical research, an international symposium was held at the Pacific Northwest Laboratory, Richland, Washington, 19–21 July 1965.

Several papers reviewed our knowledge of swine genetics and reproduction and current research in these areas. D. F. Cox (Iowa State University) reviewed swine genetics, noting that domestic swine may provide some unique opportunities for research because of the vast array of genetic variation both between and within the various breeds. He pointed out that, with the exception of information on the inheritance of blood antigens of swine, our knowledge for this species of the genetics of inherited traits that are controlled by a few genetic factors is limited. Expanding on this latter point, J. Moustgaard and M. Hesselholt (Royal Veterinary and Agricultural College, Copenhagen) reported that the presence or absence of antigenic factors on the surface of swine erythrocytes is controlled by alleles belonging to 14 chromosomal loci. Fourteen blood-group systems have been established and designated by letters.

R. A. McFeely (University of Pennsylvania) presented recent work on swine cytogenetics. The pig appears particularly well suited for studies of cytogenetics because it has 38 chromosomes, which can be paired and grouped as readily as human chromosomes.

D. Smidt and associates (University of Göttingen) have transferred the eggs from miniature swine sows to other miniature swine sows and reciprocally between miniature and Landrace sows. Embryo implantation rate was 25 percent, and about one-half of these were carried to term. The size of the sow influenced the birth weight, and the weight differential persisted until the offspring were about 6 weeks old.

Birthe Palludan (Royal Veterinary and Agricultural College, Copenhagen) presented an interesting, comprehensive review of studies on the teratological effects of vitamin A in swine with avitaminosis and hypervitaminosis. A number of malformations were observed, the most frequent being microphthalmia.

Several papers described dental and skeletal research. E. B. Jump and M. E. Weaver (University of Oregon), who pioneered in the use of miniature swine in dental research, described some advantages and limitations of this species. They noted that the pig masticates with both incision and trituration and is unique among the common laboratory animals in having a long period of deciduous and transitional dentition. The length of this period permits experimental studies on many dental problems afflicting children. Preliminary studies demonstrate the suitability of miniature swine for clinical experiments in orthodontics, periodontics, restorative dentistry, and the pathology and therapeutics of the tooth pulp. An interesting but unexplained difference between man and swine is that swine produce large amounts of dental calculus without experiencing the periodontal disorders associated with dental calculosis in man.

F. A. Spurrell, W. J. L. Felts, and L. A. Baudin (University of Minnesota) presented data on the development of osteons in swine and man and

the effect of dietary calcium restrictions. Cortical bone development in miniature pigs was found to be similar to that in the standard domestic pig.

The structure of bones of starved pigs was evaluated by C. W. M. Pratt and R. A. McCance (University of Cambridge). Newly weaned pigs were subjected to severe undernourishment for periods up to 1 year; their weights were maintained at 3 to 8 kilograms. Their bones, although showing no specific pathology, were structurally distinctive and unlike either normal growing bone or miniature mature bone. On the basis of the ratio of calcium to collagen, the bone appeared to be hypercalcified. These undernourished animals recovered when they were allowed unlimited food. All bones ultimately approached their expected length, although mild deformities of the shaft appeared in some bones.

Only one paper on renal physiology was presented. T. W. Nielsen, C. A. Maaske, and N. H. Booth (University of Colorado and Colorado State University) reported their work on standard renal-function tests performed on conscious unanesthetized pigs. Endogenous creatinine, insulin, *p*-aminohippurate, and osmotic clearances were determined. Nielsen *et al.* reported that the pig has predominantly short-looped nephrons (97 percent), compared to 86 percent for man. In the dog, however, nearly 100 percent of the nephrons are long. The ability of the pig to concentrate urine was also comparable to man; however, it appeared to be insensitive to urea loading.

Several papers discussed gastrointestinal function and nutrition. D. F. Magee (University of Washington) found that the volume of pancreatic secretion in pigs is directly related to the concentration of hydrogen ions in the duodenum from *p*H 7 to *p*H 1 and (unlike in the dog) peptone, oleic acid, and olive oil all increased the output of amylase and lipase in the juice. Above *p*H 7, secretion was inhibited. The pancreas seems to be involved in the regulation of duodenal *p*H on both sides of neutrality. Magee emphasized the advantages of using the pig rather than the dog for these studies.

W. G. Huber and R. F. Wallin (University of Illinois) analyzed the gastric secretion from swine bearing Heidenhain pouches or simple gastric fistulas. Differences were noted in *p*H, free acid, chloride, pepsin, and histamine between the two types of surgically prepared swine. However, no significant difference was observed in total secretion of acid. Difficulty was encountered in maintaining electrolyte balance in the swine with the Heidenhain pouches.

Studies on the sex-related differences in body composition in the pig and human infant were described by L. J. Filer and associates (University of Iowa). He noted that information on body composition of human infants is almost entirely restricted to that which may be accumulated by indirect methods of study. He proposed that the growing pig be used as a model for comparisons with the human infant, permitting studies on the influence of age, sex, and diet on the rate of growth and body composition during infancy.

The results of severe protein malnutrition in pigs were discussed by W. G. Pond and associates (Cornell University). They found that when pigs were weaned at 3 weeks of age to dry, low protein diets, biochemical and anatomical changes occurred which resembled kwashiorkor in human infants. When a 3-percent protein, low-fat diet was fed, the pigs remained active and alert and failed to develop the severe liver pathology and edema seen in pigs fed a 3-percent protein, high-fat diet.

B. C. Johnson (University of Illinois) also reported on the results of undernutrition of swine, with emphasis on enzymatic and cardiovascular effects of starvation and refeeding. Apparently irreversible damage was produced in myocardium as well as arteries and arterioles as a result of stresses of refeeding following starvation. Johnson noted diastolic hypertension after only two starvation-refeeding episodes.

In recent years, swine have been used increasingly in cardiovascular research. In deference to this widespread interest an evening panel was devoted entirely to cardiovascular research. D. K. Detweiler (University of Pennsylvania) reviewed the growing utilization of swine in cardiovascular studies, especially in North America and Germany. W. von Engelhardt (School of Veterinary Medicine, Hannover) presented a comprehensive review of swine cardiovascular physiology.

H. Luginbuhl (University of Pennsylvania) discussed spontaneous atherosclerosis in swine. Although it has been known for a decade that pigs develop lesions comparable to those of the preatheromatous phase in human atherosclerosis, Luginbuhl is the first scientist to report on a large number of aged swine (ranging from 8 to 14 years old). In these swine, preatheromatous changes progressed to the formation of atheroma in the aorta, iliac, cerebral, and coronary arteries of several animals. Luginbuhl identified all tissue elements constituting atheroma of man; however, ulceration and thrombus formation were the features of complicated atherosclerosis not observed so far.

H. C. Rowsell and associates (Ontario Veterinary College) found that diets high in butter, egg yolk, and lard plus cholesterol increased the amount of atherosclerosis and thrombosis in swine. They observed that, in endothelial preparations from swine as young as 2 weeks of age, deposits rich in platelets occurred around vessel orifices and bifurcations in a topography and pattern similar to early atherosclerosis. Swine were found to be ideally suited for the study of the interactions of the vessel wall, the blood platelet, and blood coagulation.

Several papers were devoted to experiments involving heart surgery. C. A. Maaske, N. H. Booth, and T. W. Nielsen (University of Colorado–Colorado State University) described experimentally induced cardiac failure in swine. By partially occluding the main pulmonary artery of the pig, they produced a slow, progressive, right-sided, congestive heart failure accompanied by signs of clinical heart failure. They noted that induction of congestive heart failure in swine was particularly significant because cardiac failure was induced by a single surgical maneuver, whereas in dogs, multiple assaults or series of surgical maneuvers are required to produce comparable results. At necropsy, grossly distended central veins, right atrial and ventricular dilatation and hypertrophy, hepatomegaly, and ascites were observed in affected animals.

G. D. Lumb (Warner-Lambert Research Institute of Canada) also reported on studies of experimentally induced cardiac failure in swine. Taking advantage of the similarity of the distribution of the coronary arteries of swine and man, he ligated the coronary branches supplying the atrioventricular node and bundle of His and then studied the degree of ensuing collateral circulation and the beneficial effects of medication.

Normal hematological and biochemical parameters of miniature swine were described by A. S. Tegeris and associates (Food and Drug Administration) and by R. O. McClellan and associates (Battelle-Northwest). They provided extensive information about more than 30 parameters. Both emphasized the need for dynamic studies in evaluating hematological and biochemical changes. McClellan emphasized the dynamic changes which appear to be age-related.

Another very interesting use of swine is in certain immunological investigations, for which they seem particularly suited. D. Segre (University of Illinois) stated that the immunological incompetence of baby pigs deprived of colostrum could be overcome by administration of antigen mixed with minute amounts of specific antibody or with large quantities of normal gamma-globulin. Such studies generated support for the natural-selection theory of antibody formation; this theory holds that the antigen-antibody complex, rather than the antigen alone, constitutes the proper antigenic stimulus. Results presented by Y. B. Kim, S. G. Bradley, and D. W. Watson (University of Minnesota) differed somewhat from those of Segre, in that they found that "immunologic virgin" pigs appear to be immunologically competent as manifested by their excellent response to antigenic stimuli—a single intraperitoneal injection of 10^{12} particles of actinophage MSP-2. They found that germ-free, colostrum-deprived miniature piglets taken by hysterectomy 3 to 5 days before term were free of detectable immunoglobulins and antibodies if great care was exercised in preventing contamination of the newborn pig with any foreign contaminant such as dam's blood. Also of related interest to immunologists was a paper by D. E. Ullrey, C. H. Long, and E. R. Miller (Michigan State University), in which they investigated the absorption of intact proteins from the first milk of the mother for protection of the newborn pig. Colostrum-deprived pigs were used to establish that intact protein could be absorbed from the intestine after birth. This was done by feeding either a protein-free or protein-containing purified diet labeled with fluorescein isothiocyanate.

F. D. Klopfer (Washington State University) reported that unless special procedures are used in rearing very young pigs, the pigs do not readily learn to solve visual discrimination problems for food reward. The effective procedures appear to be those preventing the strong development of position responses in feeding, so as to permit the development of visual discrimination while feeding. Using procedures for discrimination learning, Klopfer demonstrated wavelength discrimination in these animals and could determine the photopic and scotopic visibility functions.

Swine have been used extensively in radiobiological studies. Scientists from the U.S. Naval Radiological Defense Laboratory (in a paper presented by N. P. Page) described the recovery pattern of swine given a large sublethal dose of x-irradiation. They found the acute $LD_{50/30}$ (mid-line air dose) of 8- to 9-month-old gilts was about 400 roentgens for 1 kvp x-rays. Recovery from the effects of 238- to 265-roentgen exposures was estimated by redetermining the LD_{50} at various times after their initial exposure. The animals had recovered from 48 percent of the initial injury by 3 days and by 7 days the majority of animals appeared to have recovered completely. In fact, the data suggested some of the animals had become "radioresistant." By 20 days the LD_{50} was about 170 percent of the LD_{50} of the unconditioned animals. Studies at longer time intervals are continuing.

In another paper on the effects of external whole-body radiation, D. G. Brown (University of Tennessee) reported on studies on the late effects of swine exposed to 15 to 700 rads of mixed neutron gamma radiation from a nuclear detonation 8 years ago. About 44 percent of the irradiated swine still survive, compared to 69 percent of the controls. The increased mortality in the irradiated swine is attributed to gastrointestinal alteration and neoplasia.

R. O. McClellan reviewed the use of swine in studies of radionuclide toxicity. He described long-term studies on the effects of daily ingestion of radiostrontium, studies on the gastrointestinal absorption of plutonium and SNAP radionuclides, and metabolism and effects of plutonium deposited in the skin. The size, gastrointestinal tract, and relatively long life span of the miniature swine favor its experimental use because these criteria are important when results are extrapolated to man. Of special interest was the reported high incidence of hematopoietic tissue neoplasms in miniature swine that have ingested large quantities of strontium-90.

For many years workers have regarded the skin of swine as being very similar to human skin; it is only recently that careful, thorough, anatomical assessment has been accomplished. In a brief review of this work, W. Montagna (University of Oregon) noted that although swine skin shares some anatomical and histochemical features with that of man, it is distinctly different. The skin of both is characterized by a sparse hair coat, a thick epidermis with a well differentiated under-sculpture, a dermis that has a well-differentiated papillary body, and, most noteworthy, a large elastic tissue content. In contrast to man, the swine dermis is poorly vascularized and the sebaceous glands contain much alkaline phosphatase. In view of his observations, Montagna recommended that caution should be exercised in suggesting that there is a strong resemblance between the two skins. G. D. Weinstein (University of Miami) found the kinetics of epidermal proliferation to be similar in man and swine. He showed that cells labeled with tritiated thymidine in the epidermal basal layer had a transit time through the viable epidermis of 14 and 13 days in the pig and human epidermis, respectively. The total turnover time of pig epidermis is about 30 days, whereas in human epidermis it is 27 or 28 days.

Two papers described the results of radiation exposure of swine skin. In the first paper, J. O. Archambeau and associates (Brookhaven National Laboratory) reported on the use of swine skin for evaluating the effects of ionizing radiations. They concluded that the histology and radiation geometry of pig skin resembled the human sufficiently to warrant its use as a system for comparing effects of different types of ionizing radiations in vivo and extrapolating these results to man.

In a paper by the late L. A. George and L. K. Bustad (Battelle-Northwest) gross observation of early and late changes in swine, sheep, and rabbit skin after acute exposures to ^{32}P or ^{90}Sr plaques were described. The pattern of early response observed in swine resembled generally that described for man.

Panels discussed the laboratory management, nutrition, disease and disease control, and the development of miniature swine.

In his introductory statement, D. C.

England recalled that in 1949 the Hormel Institute of the University of Minnesota initiated a project to develop a breed of miniature swine specifically for use in biomedical research. Medical personnel of the Mayo Foundation gave added impetus, recognizing the need for a convenient experimental animal that would better meet certain anatomical and physiological demands. The objectives of the University of Minnesota program were to develop normal swine small enough at maturity to be easily handled and maintained. The objectives of the other miniature-swine developmental programs are similar and it appears that steady progress is being made in achieving these objectives.

As the programs have developed it has become clear that two classes of miniature swine need to be established. Not only is there a real demand for swine that have an adult weight less than 30 kilograms, but also for one that has an adult weight about 70 kilograms, similar to the so-called standard man and to the weight of most of the miniature swine now being used. With the development of a standard 30-kilogram pig, which may well be available during the next decade, swine will assume a major role in research laboratories. In fact, an appreciable increase in usage is predicted before this lower weight is realized.

The proceedings of the symposium will be published in book form as a publication of the U.S. Atomic Energy Commission's Division of Technical Information and will be available in early 1966 from Clearinghouse for Federal Scientific and Technical Information, U.S. Department of Commerce, Springfield, Virginia.

L. K. BUSTAD
Radiobiology Laboratory,
University of California,
Davis 95616

R. O. MCCLELLAN
U.S. Atomic Energy Commission,
Washington, D.C.

Forthcoming Events

June

25–26. **Drug Information** Assoc., annual mtg., Chicago, Ill. (E. Conrad, American Medical Assoc., Chicago)

26–28. Society for **Investigative Dermatology**, Chicago, Ill. (G. W. Hambrick, Jr., 3400 Spruce St., Philadelphia, Pa.)

26–29. American Soc. of **Agricultural Engineers**, annual mtg., Univ. of Massachusetts, Amherst. (J. L. Butt, P.O. Box 229, St. Joseph, Mich.)

26–30. American **Medical** Assoc., 99th annual mtg., Chicago, Ill. (The Association, 535 N. Dearborn St., Chicago, Ill. 60601)

26–30. American **Veterinary Medical** Assoc., 103rd annual mtg., Louisville, Ky. (The Association, 600 S. Michigan Ave., Chicago, Ill.)

26–1. American **Physical Therapy** Assoc., Los Angeles, Calif. (L. Blair, 1790 Broadway, New York 10019)

26–1. American Soc. for **Testing and Materials**, 69th annual mtg., Atlantic City, N.J. (ASTM, 1916 Race St., Philadelphia, Pa.)

26–3. National **Education** Assoc., conv., Miami Beach, Fla. (W. G. Carr, NEA, 1201 16th St., NW, Washington, D.C.)

27–28. **Astronomical** Soc. of the Pacific, annual summer mtg., Seattle, Wash. (P. W. Hodge, Dept. of Astronomy, Univ. of Washington, Seattle 98105)

27–28. **Fluorine Chemistry**, symp., Ann Arbor, Mich. (R. W. Parry, Dept. of Chemistry, Univ. of Michigan, Ann Arbor 48104)

27–29. **Aerospace Sciences**, West Coast mtg., Los Angeles, Calif. (W. J. Brunke, American Institute of Aeronautics and Astronautics, 1290 Sixth Ave., New York 10019)

27–29. **American Soc. of Heating, Refrigerating, and Air-Conditioning Engineers**, Toronto, Ont., Canada. (R. C. Cross, 345 E. 47 St., New York 10017)

27–29. **Marine Technology** Soc., 2nd annual conf., Washington, D.C. (C. W. Covey, Undersea Technology, 617 Lynn Bldg., 1111 N. 19 St., Arlington, Va. 22209)

27–29. Association for **Research in Ophthalmology**, mtg., Chicago, Ill. (H. E. Kaufman, Dept. of Ophthalmology, Univ. of Florida College of Medicine, Gainesville)

27–29. **Vacuum Metallurgy** Div., American Vacuum Soc., 9th annual mtg., New York, N.Y. (M. A. Orehoski, U.S. Steel Corp., Applied Research Laboratory, Monroeville, Pa. 15146)

27–30. **Health Physics** Soc., annual mtg., Houston, Tex. (J. G. Terrill, Jr., Div. of Radiological Health, U.S. Public Health Service, Washington, D.C.)

27–30. **Molecular Biology of Viruses**, symp., Univ. of Alberta, Edmonton, Canada. (J. S. Colter, Dept. of Biochemistry, Univ. of Alberta, Edmonton)

29–1. **Chemistry of Sulfides**, conf., Princeton Univ., Princeton, N.J. (J. Sapoch, 306 Nassau Hall, Princeton)

31–3. **Tissue Culture** Assoc., annual mtg., San Francisco, Calif. (W. A. Nelson-Rees, Naval Biological Laboratory, Naval Supply Center, Oakland, Calif., 94625)

July

1–3. **Radiology of Normal and Pathological** Mammary Structures, European symp., Strasbourg, France. (C. Gros, Service Central de Radiologie, Hôpital Civil, Strasbourg 67)

4–8. **British Medical Assoc.**, Exeter, England. (Secretary, Tavistock Sq., London W.C.1, England)