BOVINE REPRODUCTIVE HEALTH IN THE 21st CENTURY

R.H. BonDurant
Dept. of Reproduction, School of Veterinary Medicine
University of California, Davis
Davis, CA 95616

Introduction

Predicting the future is a skill that is not taught in most veterinary curricula, and not practiced by most learned veterinarians. It is an endeavor fraught with hazard, the greatest of which being that a small but wicked minority of those who hear one's predictions will remember them at an importune moment in the future.

Nevertheless, I could not resist accepting the invitation to come to the World Buiatrics Meeting, in order to expound on the subject of bovine reproductive health in the 21st Century. There were several compelling reasons to accept the invitation, namely:

1. Minnesota is lovely in the late summer.
2. The Twins may be in town.
3. I am just barely below the median age of the assembled group, and can perhaps outlive the wickedness of those few who may remember what I say here.

I accepted the offer to look into the future with a profound sense of humility, which I will now proceed to justify. While my overwhelming inner response to the suggested title for this presentation was "How on Earth should *I* know what bovine reproductive health will be like in the next century?", I quickly became intrigued by:

a. The prospects of following current technological progress to its logical sequelae;
b. The possibilities of integrating technologies from different disciplines;
c. The idea that in my lifetime, we may discover the cause, treatment and prevention of various disease processes that have eluded us for decades in *this* century.

However, being intrigued and being inspired are two different things. I waited patiently for the appropriate muse to enlighten me. I even tried invoking him (her?) by reading "leading" journals and watching "futuristic" special presentations on television. I consulted with colleagues, most of whom, like me, have difficulty predicting the outcome of the current *day*, much less the coming century. None of this invocation was of any particular use, so I tried a new tactic: I consulted the past. Whether the cliche that the "past is prologue" is true or not, it seemed to me that I might at least gain an appreciation of how we got "here," so that getting "there" might seem less mysterious.

The Twentieth Century

Veterinary medicine in general, and animal reproduction in particular, came of age in this century. After untold centuries of using parturition as proof of pregnancy (the biological equivalent of testing a firecracker), a practical means of early diagnosis of pregnancy was established. While the precise origin of the technique of trans-rectal palpation is not well documented, it is clear that the technique was mentioned in early twentieth century veterinary literature (4,6,8,17,18, and others). A body of literature was compiled

and interpreted by Fleming in 1906 (6), and included most of the pertinent 19th century wisdom of authors from France, Germany, Belgium, Holland, Sweden, Austria, Poland, Italy and England. It is interesting to note that Fleming and others were not afraid to offer some opinions along with their objective observations, e.g.:

> "...various expedients have been resorted to in order to ascertain with certainty the presence of the foetus in the uterus, by exciting ...movements. Some of these are as barbarous as they are stupid. One especially merits the strongest condemnation; this is pouring water into the ears of the Mare or Cow, under the supposition that if the animal is not pregnant it will shake its whole body to get rid of the fluid, but if pregnant that it will only shake the head and ears..."

Most of the early authors advocated trans-rectal methods of determining bovine pregnancy after the pregnancy was well established, i.e. at or after the third month of gestation. Ultimately, a reliable method for early detection of pregnancy, namely the palpation of a chorio-allantoic "membrane slip" or the detection of an amniotic vesicle, became popular, although formal written descriptions of it are rare before the 1940's (3).

Today, many decades after the description of these techniques, trans-rectal palpation of early pregnancy remains the cornerstone of veterinary reproductive management programs. The importance of the technique reflects the relative importance of reproductive performance in the overall success of the livestock enterprise. While other newer technologies exist that can either suggest or strongly suggest pregnancy without palpation, it is clear that they do not match palpation by a qualified veterinarian in terms of cost-effectiveness.

Other accomplishments of this century include: the biochemical and physiological characterization of the major reproductive hormones; the exploitation of this endocrine knowledge to allow for clinical diagnostic assays (including exquisitely sensitive "cow-side" assays), estrous cycle control, superovulation, therapeutic luteolysis, induction of parturition, etc.; the development of practical methods of artificial insemination, including one of the most significant contributions of the century, the ability to successfully freeze semen; the development of surgical and then non-surgical means of embryo collection and transfer; the manipulation of embryos to produce multiple individuals from a single embryo (embryo splitting, embryo cloning), or a single embryo from multiple individuals (chimerism); and the insertion of exogenous genetic material into the fertilized egg.

We have witnessed the beginnings of answers to the critical question of how the dam recognizes that she is pregnant. We have also seen the reduction of the impact of many infectious disease on reproductive performance, thanks largely to effective diagnostic, eradication and immunoprophylactic strategies. And as the century draws to a close, we are beginning to see a new picture of the means by which the reproductive tract defends itself from pathogens.

Given all these technical achievements, it is logical to ask: Will veterinarians - who have all this technology to draw from - still be palpating cows well into the 21st century? A significant body of veterinarians hopes not. That is, the profession has come so far in technical, medical and management skills that many would be willing to abandon the "green arm" image of the profession in favor of something more sophisticated. So let me depart from the limitations of today, and wander shamelessly into the realm of fantasy, where technical advances may offer some relief for the profession's weary left (or right) arm.

Reproductive management in tomorrow's milking parlor

Imagine a milking parlor, plumbed much like the more modern parlors of today, except that the milking unit is not applied to the cow by a human, but rather by a robot (Ford Motor Company or

Mitsubishi could show us how). The cow is of course identified electronically, and her identity is reported to the on-board mini-computer in the dairyman's office. Decisions about the disposition of individual cows at each "station" are transmitted from the computer to the robot (e.g. if yesterday's production qualifies her to move to a lower production string, the appropriate gate movements will direct her out of the parlor after milking). A micro-transmitter installed intravaginally transmits information to the computer about the electrical conductivity of the vaginal mucus of each cow.

When the milking machine is robotically applied, the following parameters are measured by the unit:

1. Milk volume (as is done today).
2. Milk temperature and electrical conductivity (can be done today).
3. Somatic cell count, in a parallel holding chamber.
4. Milk urea levels, so that the potential negative influence of high protein diets can be minimized.
5. Milk fat percentage and milk ketone levels.
6. Milk protein percentage.
7. Reproductive hormone levels.

As to which hormones to measure, the picture is not completely clear at the moment. Progesterone is a logical choice, as there are already colorimetric assays for its measurement which might integrate well with a mechanical system as described above. But the limitations of progesterone interpretation present a problem: what does an elevated progesterone mean, and how much should we believe it? What about other hormones, like bovine pregnancy-specific protein B (bPSPB)? This hormone, derived from binucleate cells of the conceptus, would surely be a more specific indicator of pregnancy than progesterone. Unfortunately, bPSPB isn't secreted into the milk in any measurable quantity (Dr. Jim Butler, U. of Idaho, personal communication), so its application in the "21st century parlor" is doubtful. We may have to await the discovery of a pregnancy-specific hormone (preferably a fat-soluble one) that is secreted into the milk before the above fantasy can be realized.

But even if we do find such a hormone, and can measure it in the system described, we will need an entirely new type of computer software in order to manage the information that will otherwise bury us. As a trivial example, one may not be too interested in the precise level of ~hormone X" (e.g. progesterone) that cow 1302 showed today, but rather in how today's level relates to the level she demonstrated over the past 5 (7,14,21, etc) days. Essentially, we want software that can interpret which cows are performing as expected, and which ones are exceptions to such performance. Such software will have to take the huge burden of interpretation of millions of pieces of information, and translate that into "exception lists" from which the manager can take action.

Many components of the above milking parlor are already technologically possible, although I'm not aware of anyone who has put them together in a single package.

Other Technologies

Dr. Phil Senger has already given notice to the livestock industry of reproductive technologies to come, including the use of sexed semen, automated estrus detection, and perhaps "timed-release" insemination (15). As with the parlor of the future, each of these technologies has been shown to be feasible in laboratory settings, and each is a long way from practical applications.

Infectious Disease: the future is not in antibiotics

This is the safest prediction one can make. It seems inevitable that the day of the intrauterine infusion of antibiotics will be regarded in the 21st century the same way we view Dr. Fleming's colleagues' "water

in the ear" method of pregnancy detection from the last century. Just exactly what will replace antibiotics is less certain. Advances in our understanding of the natural immune defense mechanisms of the cow's reproductive tract, and perhaps our ability to genetically or therapeutically enhance them, will probably reduce or eliminate our dependence on compounds that the public views as a threat to its food supply.

It seems logical to project that further work in the area of uterine immunity will help us understand the reproductive tract's ability to distinguish between conceptus and pathogen, and allow us to capitalize on that distinction. Examples include the enhancement of specific subsets of uterine lymphocytes, or the purification and therapeutic use of natural microbicidal peptides from host leukocytes (2). Other attractive research targets include cytokines, chemoattractants, endotoxins, and serum opsonins (5,7). It is even possible that we may administer vaccines directly to the reproductive tract, e.g. *A. pyogenes* (or a fraction thereof), in order to stimulate an effective local immunity. Similarly, we may administer passive immunity directly into the vagina for protection against venereal agents, using time-release technology, as has been shown for the mouse (13).

We will probably see the application of recent molecular biological techniques which will allow us to bypass Koch's postulates in establishing etiologies. Diseases whose etiology has eluded us for decades may be "revealed" by analysis of nucleic acids of microbes, even when the whole microbe cannot be grown in culture. Such techniques can accurately place the causative organism into its appropriate place on the phylogenetic tree. They have been applied already in human disease (11), and may be of significant help in bovine disease next century.

Perhaps we will routinely use paracrine products of the conceptus as therapeutic aids for reducing the amount of embryonic death that occurs due to failure of maternal recognition of pregnancy (1, 9,12,14,16). These products, the interferon-like bTP-1 and other immunosuppressive molecules, are already being cloned (10), so that large amounts may someday be available for therapy.

What kind of animals?

The "standard equipment" twenty-first century cow will probably look familiar to most of us. But her genes may include a few "options" that have been artificially added. Whereas today we are familiar with the effects of repeated administration of recombinant somatotropin on milk production, are we ready for the cow that has had the gene(s) for somatotropin added to her own complement of genes? Such a cow could theoretically be produced today (it's been done in sheep); but we still need to know much, much more about the control of such genes before the BST supercow is a reality.

Nevertheless, other transgenic animals will appear quite early in the 21st century. Cows whose milk components have been transgenically altered will be available, such that we may have herds of cows producing low-fat milk (skeptics would say we've already done that), or cows with genes for particular casein types, so that cheese yields are increased. We will probably also see an increase in the use of cows for the production of useful pharmaceuticals, excreted in the milk. As with other "futuristic" technologies, this one is being done now, in small scale settings.

We will be able to select animals for disease resistance (to specific pathogens), based on their genetic immunological makeup, i.e. based on their major histocompatibility (MHC) profiles. Currently, researchers are looking for associations between MHC types and susceptibility to diseases of international economic importance; but we may get to the point where resistance to locally important "nuisance diseases" may be selected against, e.g. *A. pyogenes* endometritis, BVD embryonic and fetal loss, etc.

What kind of Veterinarian?

While we're on the topic of genetics, it is obvious that the chromosomal constitution of next century's veterinarians will be statistically different from those of this century...that is, the proportion of the livestock veterinary population that sports a Y chromosome will be significantly lower ($P<0.05$) than it has been this century. This is not only a welcome change, it is inevitable: The trend towards the feminization of our

profession has been irreversible for the last 20 years, and shows no signs of ceasing. As a profession, we need to ensure that the livestock veterinarian of the 21st century is representative of the best and brightest of the classes we are graduating now, which means there will be plenty of women serving our cattle industries.

Aside from a gender shift, our veterinarians will have to be better trained than we were. They will need to have a foundation in the science of animal husbandry; in theoretical and applied bovine nutrition; in animal agricultural economics; in facilities design; in computer usage for day-to-day functions and for periodic analytical functions; in interpersonal skills; in people management; in data management; in problem solving...and maybe, if they've mastered all of these, in palpating a cow.

Summary

The 21st century will see an increase in the incorporation of biotechnology into the day-to-day operation of reproductive health programs for cattle. Veterinarians, an increasing number of whom will be women, will be called upon to interpret biological and management data, to examine performance, and to scientifically test new strategies for enhancing that performance.

References

1. Arthur, G.H., Noakes, D.E., Pearson, H., (1989). *Veterinary Reproduction and Obstetrics (Theriogenology)*, 6th ed. Balliere and Tindall, London; p 113. 2. Boernstein, L.A., Ganz, T., Sell, S., Lehree, R.I., Miller, J.N., (1991). Contribution of rabbit leukocyte defensins to the host response in experimental syphilis. *Inf Imm* 59(4):1368-1377. 3. Cowie, A.T., (1948). Pregnancy diagnostic tests: a review. *Grt Brit CAB joint publication no.* 13:11-15. 4. Craig, J.F., (1930). *Fleming's Veterinary Obstetrics*, 4th ed. Balliere, Tindall and Cox, London. 5. Cullor, J.S., Wood, S., Smith, W., Panico, L., Selsted, M.E., (1991). Bactericidal potency and mechanistic specificity of neutrophil defensins against bovine mastitis pathogens. *Vet Microbiol* 29(1):49-58. 6. Fleming, G., (1906). A Textbook of Veterinary Obstetrics, 2nd ed. W.R. Jennings, New York. pp 142 ff. 7. Ganz, T., Selsted, M.E., Lehrer, R.I., (1990). Defensins. *European J. Haematology* 44(1):1-8. 8. Hess, E., and Joss, E., (1921). Die Sterilitat de Rindes. M. & H. Schaper, Hannover, pp 92 ff. 9. Newton, G.R., Vallet, J.L., Hansen, P.J., Bazer, F.W., (1989). Inhibition of lymphocyte proliferation by ovine trophoblast protein-1 and a high molecular weight glycoprotein produced by the peri-implantation sheep conceptus. *Am J Reproductive Immunology* 19(3):99-107. 10. Ott, T.L., Van Heeke, G., Johnson, H.M., Bazer, F.W., (1991). Cloning and expression in Saccharomyces cerevisiae of a synthetic gene for the type-1 trophoblast interferon ovine trophoblast-1 protein: purification and antiviral activity. *J Interferon Res* 11(6):357-64. 11. Relman, D.A., Loutit, J.S., Schmidt, T.M., Falkow, S., Tompkins, L.S.,(1990). The agent of bacillary angiomatosis. *New Eng J Med* 323(23):1573-80. 12. Roberts, S., (1986). Veterinary Obstetrics and Genital Diseases (Theriogenology). Published by the author, Ithaca, New York, p 17-18. 13. Rodomsky, M.L., Whaley, K.J., Cone, R.A., Saltzman, W.M., (1992). Controlled vaginal delivery of antibodies in the mouse. *Biol Reprod* 47(1):133-140. 14. Segerson, E.C., and Bazer, F.W., (1989). High molecular weight basic and acidic immunosuppressive protein components in uterine secretions of pregnant cows. *Biol Reprod* 41(6):1014-23. 15. Senger, P., (1992). Reproductive management in the future. *Hoard's Dairyman* 137(9):388. 16. Thatcher, W.W., Hansen, P.J., Gross, T.S., Helmer, S.D., Plante, C., Bazer, F.W., (1989). Antiluteolytic effects of bovine trophoblast protein-1. *J Reprod Fert* Supplement, 37:91-99. 17. Williams, W.L., (1940). Veterinary Obstetrics, 3rd ed. Published by the author, Ithaca, New York, pp 113-114. 18. Williams, W.L., and Williams, W.W., (1921). The Diseases of the Genital Organs of Domestic Animals. Published by the authors, Ithaca, New York.