

was investigated during a three-year study in the Etosha National Park on five lion prides. ...Individuals were used as control animals or given melengestrol acetate...implants, branded and released; the animals were observed for possible changes in behavior, birth rate and mortality. Ten treated lionesses were recaptured at irregular intervals to reassess weight and steroid hormone levels, while three lionesses had their implants removed to determine if their fertility would return. The contraceptives prevented pregnancy, were reversible when removed and did not alter lion behavior significantly, except that sexual behavior was not recorded in treated lionesses.

Paterson, M., and R. J. Aitken. 1990. Development of vaccines targeting the zona pellucida. *Current Opinion in Immunology* 2:743-747.

IMMUNOCONTRACEPTION, OVERVIEW, ZONA PELLUCIDA

A good, though moderately technical, overview of research on the zona pellucida as a target for contraceptive vaccines. The focus is towards developing a vaccine for use in humans, but most of the paper summarizes research using animal models.

Regarding the mechanism involved in contraception:

"Anti-ZP antibodies are thought to exert their contraceptive effect in one of two ways: by direct interaction with the sperm receptor site, or by binding to antigens that are close to the receptor site, masking the sperm receptor by steric hindrance." Furthermore, anti-ZP antibodies may hinder the reproductive process either by blocking folliculogenesis or the sperm-egg interaction.

The review also points out some of the problems (side effects) which have characterized past studies, due at least in part to impurities in the vaccines.

The paper also contains a brief list of annotated references.

"Recent publications have emphasized the importance of identifying the epitopes on the surface of the ZP that are uniquely involved in the fertilization process. A potential vaccine should involve antibodies that inhibit fertilization by preventing sperm binding to and penetrating the oocyte without causing ovarian dysfunction. From the literature, it would seem that the use of highly purified well defined epitopes as immunogens is a significant step in this direction. ...With a knowledge of the precise antigenic determinants involved in fertilization, and their mechanism of action, it should be possible to engineer vaccines in which the disturbance of ovarian function is minimized or eliminated." (p. 747)

Paterson, M., P. T. Koothan, K. D. Morris, K. T. O'Byrne, P. Braude, A. Williams, and R. J. Aitken. 1992. Analysis of the contraceptive potential of antibodies against native and deglycosylated porcine ZP3 in vivo and in vitro. *Biology of Reproduction* 46:523-534.

FEMALE, IMMUNOCONTRACEPTION, MONKEY, PZP, ZONA PELLUCIDA

Abstract

We have undertaken a comparative analysis of the contraceptive activity of antibodies directed against the porcine sperm receptor zona pellucida antigen (ZP3) and its $M_r=32\ 000$ polypeptide core (DGZP-32). The strategies employed for this analysis included the induction of active immunity in a primate, the common marmoset, and an in vitro fertilization protocol involving the use of viable human ova. In both experimental situations, antibodies against ZP3 were shown to exhibit contraceptive activity, leading respectively to the induction of long-term infertility in the primate model and to the complete inhibition of human fertilization

in vitro. The in vivo studies also revealed that the induction of high titer antibodies against ZP3 were inevitably associated with the appearance of an ovarian pathology characterized by the progressive depletion of the primordial follicle pool within one to two years. This side effect could not be alleviated by the use of DGZP-32 as an antigen since the induction of immunity against this polypeptide was also associated with the eventual appearance of an ovarian pathology identical to that observed with ZP3. Furthermore, the DGZP-32 peptide was less effective than ZP3 in inducing the formation of antibodies capable of inhibiting the fertilization of human ova in vitro. We conclude that significant problems remain with the use of deglycosylated zona peptides for the development of contraceptive vaccines and that their potential will not be realized until the epitopes responsible for the induction of infertility and the primordial follicle depletion have been identified and segregated.

Pelletier, G., L. Cusan, C. Auclair, P. A. Kelly, L. Desy, and F. Labrie. 1978. Inhibition of spermatogenesis in the rat by treatment with [D-Ala,Des-Gly-NH₂]LHRH ethylamide. *Endocrinology* 103:641-643.

CLINIC, LHRH, MALE, MAMMAL, RAT

Abstract

The effect of treatment with a potent LHRH agonist, [D-Ala, Des-Gly-NH₂¹⁰LHRH ethylamide, injected at the low dose of 100ng, twice a week, was evaluated on spermatogenesis in the rat. Significant degenerative changes of seminiferous tubules could be observed after two weeks of treatment. These changes were progressive and led to a marked inhibition of spermatogenesis after four to eight weeks of treatment. Testis weight was decreased to approximately 50% of control after eight weeks of treatment.

Pineda, M. H., L. C. Faulkner, M. L. Hopwood, and D. C. Lueker. 1968. Effects of immunizing female rabbits with bovine luteinizing hormone. *Proceedings of the Society for Experimental Biology and Medicine* 128:743-749.

CLINIC, FEMALE, IMMUNOCONTRACEPTION, LHRH, MAMMAL, RABBIT

"Injections of purified bovine LH...in rabbits produced circulating antibodies which were associated with degenerative changes in uterine, ovarian, and adenohipophyseal structures and affected mating behavior and ovulation. Final body weight; gain in body weight; adrenal, thyroid, and thymus weights; and histology were unaffected. Freund's complete adjuvant caused microscopic changes in the ovaries and adenohipophysis. Bovine LH and crude rabbit anterior pituitary extract contained a similar antigen which cross-reacted with anti-BLH serum. The cross-reacting antigen was not bovine gamma globulin or bovine serum albumin. Results were attributed to an autoimmune phenomenon with circulating anti-bodies common to [bovine LH] and endogenous gonadotropin."

Plotka, E. D., and U. S. Seal. 1989. Fertility control in female white-tailed deer. *Journal of Wildlife Diseases* 25:643-646.

CLINIC, DEER, FEMALE, IMPLANT, MAMMAL, MELENGESTROL ACETATE

Abstract

Silastic rods containing either melengestrol acetate (MGA) or levonorgestrel (LN) were placed in anestrous white-tailed deer (*Odocoileus virginianus borealis*) does to evaluate the contraceptive efficacy of the implants over a 2 yr period. Implants of MGA were placed in five does during mid-pregnancy to evaluate the effect of this treatment on pregnancy, parturition and lactation. Pregnancies were not observed in the

five animals implanted with MGA during anestrus. Three of five does implanted with LN became pregnant in the first season. Pregnancy was not interrupted in the five pregnant does implanted with MGA and it was necessary to remove the implants and treat the does with an estrogen to achieve parturition. One of five fawns was delivered alive and was raised by the doe. MGA was effective for 2 yr as a contraceptive in white-tailed deer, LN was ineffective as used, and MGA placed in pregnant does delayed or prevented normal parturition and thus should not be used in pregnant deer.

Plotka, E. D., T. C. Eagle, D. N. Vevea, A. L. Koller, D. B. Siniff, J. R. Tester, and U. S. Seal. 1988. Effects of hormone implants on estrus and ovulation in feral mares. *Journal of Wildlife Diseases* 24:507-514.

CLINIC, FEMALE, HORSE, IMPLANT, MAMMAL, STEROID

Abstract

Five groups of 30 captive feral mares each were implanted with silastic rods containing estradiol...progesterone...[or some combination of these drugs]. ...In spite of the significant effect of hormone implants on the occurrence of estrus, over 80% of mares bred and conceived when placed with a stallion. ...These hormones, when implanted during early anestrus, can alter the occurrence of psychic estrus in captive feral mares without suppressing ovulation and conception.

Plotka, E. D., D. N. Vevea, T. C. Eagle, J. R. Tester, and D. B. Siniff. 1992. Hormonal contraception of feral mares with silastic rods. *Journal of Wildlife Diseases* 28:255-262.

CLINIC, FEMALE, HORSE

Abstract

Homogeneous Silastic(R) rods containing ethynylestradiol (EE) (1.5 or 4g), estradiol-17 β (E) (4g) or progesterone (P) (6g) were implanted into feral mares (*Equus caballus*) between 4- and 10-yr-old. ...All animals pregnant at the time of contraceptive placement delivered normal foals. Contraceptive efficacy for groups LEE, MEE, HEE and P+HEE were 75, 75, 100 and 100%, respectively after two breeding seasons. Suppression of ovulation appeared to be inversely related to the concentration of EE used in the implant. ...Contraceptive efficacy was followed for 3 yr in one group, P+HEE, and was 88%. ...Our data demonstrate effective contraception of feral mares for up to 36 mo without compromising a pregnancy in effect at the time of implanting. Calculating the decline in EE concentrations to 150% of pre-implantation concentrations, these data suggest an effective contraceptive life of approximately 16, 26, and 48 to 60 mo for LEE, MEE, and HEE, respectively. Mechanisms that appear to be involved in contraceptive efficacy include preventing ovulation at higher concentrations of steroids and either suppressing ovulation or implantation at lower concentrations of steroid. ...

Porton, I., C. Asa, and A. Baker. 1990. Survey results on the use of birth control methods in primates and carnivores in North American zoos. Pages 489-497 in AAZPA 1990 Annual Conference Proceedings.

CHEMOSTERILANT, CLINIC, MAMMAL

Reports on the results of two surveys distributed to 105 (re: primates) and 113 (re: carnivores) zoos in North America. Response rates were 61.9% and 53%, respectively.

This report contains a summary of birth control methods and the problems or success that zoos have had.

Tables include convenient encapsulation of reported methods and reversibility of techniques.

Portugal, M. M., and C. S. Asa. 1995. Effects of chronic melengestrol acetate contraceptive treatment on perineal tumescence, body weight, and sociosexual behavior of Hamadryas baboons (*Papio hamadryas*). *Zoo Biology* 14:251-259.

BABOON, CLINIC, FEMALE, IMPLANT, MAMMAL, MELENGESTROL ACETATE

Abstract

Melengestrol acetate (MGA) subcutaneous implants were used in seven adult female Hamadryas baboons to study the effects of chronic progestin administration on perineal swelling, behavior, and weight gain. At the dose level used (≥ 8 mg/kg body weight), cycles of perineal tumescence ceased and swellings subsided. Social dynamics were not significantly altered during treatment, and only minor changes in agonistic interactions were noted following implant removal. Body weight increased by 17.3-47.6% during treatment despite decreased time spent foraging. This paradox may be explained by 1) foraging time not necessarily corresponding to total intake and 2) the nonsignificant trend toward lower levels of activity during treatment. Treatment with MGA can be recommended based on its ability to suppress ovarian cyclicity without causing social disruption. However, because this and other progestins stimulate weight gain, their use is contraindicated in overweight or obese animals.

Potvin, N. J., J. M. Bergeron, M. Norman, and A. Cyr. 1982a. Evaluating the sterile male method on red-winged blackbirds: clinical evaluation of thiotepa as a sterilant. *Canadian Journal of Zoology* 60:460-465.

AVES, CHEMOSTERILANT, CLINIC, MALE, RED-WINGED BLACKBIRDS, THIOTEPA

Abstract

Before attempting a small-scale field trial of the sterile male method to reduce blackbird population levels, laboratory studies were undertaken, in 1978 and 1979, to evaluate the efficiency of thiotepa as a bird sterilant. The LD50 of thiotepa was determined (6.2 mg/kg of body weight), as well as the applicable dose to produce sterility (4.32 mg/kg of body weight) without causing mortality, following a treatment beginning on April 25, 1979. This dose inhibited testicular growth for about 1 month. During this period, testes weight of treated redwings was only 10% of normal weight. Identical doses to those administered to male redwings produced the same effects on testicular development of grackles and cowbirds. No effects were noted on the testes weight of male Red-winged Blackbirds 1 year after treatment with thiotepa. Results indicated that the sterile male approach may have potential to reduce blackbird populations causing crop damage.

Potvin, N. J., J. M. Bergeron, M. Norman, and A. Cyr. 1982b. Evaluating the sterile male method on red-winged blackbirds: effects of the chemosterilants thiotepa on the reproduction of clinically treated birds under field conditions. *Canadian Journal of Zoology* 60:2337-2343.

AVES, CHEMOSTERILANT, CLINIC, MALE, RED-WINGED BLACKBIRDS, THIOTEPA

Abstract

In 1978 and 1979, spring blackbird flocks composed of Red-winged Blackbirds, Common Grackles, and

Brown-headed Cowbirds reached a maximum of 5000 birds in the study area. Red-winged Blackbirds accounted for a majority of birds in these flocks. Flocking lasted approximately 1 month leaving ample time for a field application of the chemosterilization method. Three sets of experiments were used to examine the feasibility and the potential of the sterile male method to reduce productivity of Red-winged Blackbird populations. Birds treated by gavage at a rate of 3.1 mg thiotepa/kg body weight had a hatching rate of 32.7%. This was significantly lower than that found in control groups (79.8 to 90.0%). The second treatment, which consisted of feeding redwings with treated corn for 4 consecutive days, thus causing a mean daily ingestion of 3.58 mg thiotepa/kg body weight, produced a hatching rate of 55.6%. The third treatment, like the second but lasting 10 consecutive days, resulted in a daily ingestion by blackbirds of 4.01 mg thiotepa/kg body weight. The hatching rate of this latter group was 46.3% and was significantly lower than that of the control group. Red-winged Blackbird testes weight remained stationary in the treated group for over 1 month after the beginning of the treatment. These experiments demonstrate that the sterilization approach in field conditions offers hope of reducing populations causing crop damage if proper field application methods are developed.

Primakoff, P., W. Lathrop, L. Woolman, A. Cowan, and D. Myles. 1988. Fully effective contraception in the male and female guinea pigs immunized with the sperm protein PH-20. *Nature* 335:543-546.

FEMALE, GUINEA PIG, IMMUNOCONTRACEPTION, MALE, MAMMAL, PH-20

Abstract

Immunization of male and female animals with extracts of whole sperm cells is known to cause infertility. Also, men and women who spontaneously produce antisperm antibodies are infertile but otherwise healthy. Although the critical sperm antigens are unknown, these observations have led to the proposal that sperm proteins might be useful in the development of a contraceptive vaccine. The guinea pig sperm surface protein PH-20 is essential in sperm adhesion to the extracellular coat (*zona pellucida*) of the egg, a necessary initial step in fertilization. Here, we report that 100% effective contraception was obtained in male and female guinea pigs immunized with PH-20. Antisera from immunized females had high titres, specifically recognized PH-20 in sperm extracts, and blocked sperm adhesion to the egg *zona pellucida in vitro*. The contraceptive effect was long-lasting and reversible: immunized females, mated at intervals of six to fifteen months after immunization, progressively regained fertility.

Remfry, J. 1978. Control of feral cat populations by long-term administration of megestrol acetate. *Veterinary Record* 103:403-404.

CAT, FIELD, MAMMAL, MEGESTROL ACETATE

A brief note reporting on a 10 month long effort to treat a small (n=14) population of free ranging house cats. The experimental design has numerous flaws which limit inferences about the outcome, which was sufficiently ambiguous anyway.

Problems related to applying this technique on a broader scale include the controlled dosage requirement, the labor required to carry out the dosing, and the apparent potential side effects of the drug compound.

Rivier, C., J. Rivier, and W. Vale. 1979. Chronic effects of [D-Trp,Pro-NEt] luteinizing hormone-releasing factor on reproductive processes in the male rat. *Endocrinology* 105:1191-1201.

CLINIC, LHRH, MALE, MAMMAL, RAT**Abstract**

Chronic administration of a potent LRF agonist, [D-Trp,Pro-NEt]LRF (A), to intact adult male rats induced a dose-related decrease in testicular and accessory organ weights, diminished production testosterone...and disrupted tubular morphology, indicative of suppression spermatogenesis. ...

Robbins, J. 1987. Seeking ways to put wild horses on the pill. High Country News 19(5,16 March):12.

FIELD, HORSE, OVERVIEW

A brief article describing efforts by Kirkpatrick and Turner to contracept herds on BLM lands.

Roberts, A. J., and J. J. Reeves. 1989. Reproductive and endocrine changes in ewes actively immunized against luteinizing hormone. Journal of Reproductive Immunology 16:187-197.

CLINIC, FEMALE, IMMUNOCONTRACEPTION, LHRH, MAMMAL, SHEEP**Abstract**

The objective of this study was to examine reproductive and endocrine changes in ewes actively immunized against a conjugation of luteinizing hormone (LH) and ovalbumin (LH immunized n=10) or ovalbumin alone (control; n=10). Ewes were immunized at weeks 0, 3 and 5 of the 93 week study and exposed to a fertile ram during weeks 7-25 (first breeding season) and weeks 60-67 (second breeding season). Immunization against LH abolished estrus and prevented pregnancy in all but one LH immunized ewe during the two breeding seasons. Uterine weights of LH immunized ewes were lighter than those observed in control ewes while ovarian weights did not differ between the treatment groups. No differences were observed between basal circulating concentrations of LH measured by an in vitro bioassay for the two treatment groups. However, serum concentrations of immunological follicle stimulating hormone, and the number of medium sized ovarian follicles (4-6mm) were increased in LH immunized ewes. It was concluded from this study that immunization against LH prevents ovulation presumably by blocking the preovulatory surge of LH and not altering basal concentrations of LH.

Roldan, E. R. S., T. Murase, and Q. X. Shi. 1994. Exocytosis in spermatozoa in response to progesterone and zona pellucida. Science 255:1578-1581.

BICUCULLINE, CLINIC, MAMMAL, MOUSE, STEROID**Abstract**

Exocytosis in mammalian spermatozoa (the acrosome reaction) is a process essential for fertilization. Both progesterone and zona pellucida induce exocytosis in spermatozoa, which may encounter both during penetration of the oocyte's vestments. When mouse spermatozoa were exposed first to progesterone and then to zona pellucida, exocytosis was enhanced to a greater degree than that seen when the agonists were presented together or in the inverse order, which suggests that the steroid exerts a priming effect. Progesterone similarly primed the generation of intracellular messengers evoked by zona pellucida. The effects triggered by progesterone were mimicked by gamma-aminobutyric acid (GABA) and were blocked by bicuculline, which indicates that the steroid acts on a GABA_A receptor.

Roughton, R. D. 1979. Effects of oral melengestrol acetate on reproduction in captive white-tailed deer. *Journal of Wildlife Management* 43:428-436.

CLINIC, DEER, FEMALE, MALE, MAMMAL, MELENGESTROL ACETATE

Abstract

Captive white-tailed deer (*Odocoileus virginianus*) of both sexes were given an antioviulatory drug, melengestrol acetate (MGA; 6 α -methyl-6-dehydro-16-methylene-17-acetoxyprogesterone), in feed at 0.6 to 1.0 mg per head daily during the October-March breeding season or for up to 12 consecutive months. MGA inhibited reproduction in adult breeding groups without adverse side effects; did not influence subsequent reproduction in such groups; and did not inhibit the growth, sexual maturation, or subsequent fertility of male or female yearlings or fawns. Discontinuation of MGA treatment of mature females in mid-December was followed shortly by ovulation and conception, but treatment during any stage of gestation from about day 1 through day 192 did not interrupt pregnancies, change gestation length, cause still birth, or induce parturition difficulty. Feed intake was not significantly influenced by the addition of MGA. Oral MGA would be feasible for regulating reproduction in small confined herds of deer, but seems impractical for application to larger free-ranging populations because antioviulatory effects require daily ingestion. Implanted MGA, other long-term sterilization treatment, or an oral gametocide appears more attractive for managing free-ranging populations.

Rowe, F. P., and A. B. Lazarus. 1974a. The effects of an oestrogenic steroid on the reproduction of wild rats, *Rattus norvegicus* (Berk.). *Agro-Ecosystems* 1:57-68.

MAMMAL, RAT, STEROID

Abstract

A dense population of rats, *Rattus norvegicus* (Berk.), infesting a refuse tip, was fed plain cereal bait for 3 weeks and then the same bait treated at 0.05% with a synthetic oestrogen, the 3-cyclopentyl ether of 17 α -hexa-1', 3'-diynyloestra-1,3,5(10)-trien-17 β -ol (BDH 10131) for 6 days.

Rats were sampled by trapping immediately before the treatment and at 4-10 weekly intervals afterwards over the next 56 weeks. Similar samplings were done at a second tip, used as a control, over a period of 32 weeks. The trapping data indicated that the reproductive potential of the treated rats was severely impaired. Sub-adult animals of either sex were not caught later than 12-weeks post-treatment, the average body-weight of the adults trapped tended to increase at successive samplings and trapping success fell. In contrast, sub-adults were found in each sample taken at the control tip and trapping success remained fairly constant throughout.

Marked differences between the treated and the control rats were found at autopsy. A majority of the adult males sampled in the 16-week period after the treatment had atrophied testes laying in the abdominal position. Their fertility of the treated adult females was also seriously affected. No successfully breeding females were caught until 26-weeks post-treatment. At the control tip most of the adult males caught had scrotal testes and pregnant females were present in each sample.

The results of the BDH 10131 treatment are discussed in relation to similar field studies done using two other synthetic oestrogens, mestranol and quinoestrol, and to practical aspects.

Rowe, F. P., and A. B. Lazarus. 1974b. Reproductive activity in a wild rat, *Rattus norvegicus* (Berk.), population treated with an oestrogenic steroid. *Agro-Ecosystems* 1:227-235.

BDH 10131, FIELD, MAMMAL, RAT, STEROID**Abstract**

A population of rats, *Rattus norvegicus* (Berk.), infesting a refuse tip was sampled by removal trapping on seven occasions over a 32-week period and then treated with a synthetic oestrogen reproduction inhibitor, the 3-cyclopentyl ether of 17 α -hexa-1', 3'-diynyoestra-1,3,5(10)-trien-17 β -ol (BDH 10131). The compound was included at 0.05% in an oatmeal-based bait and the treated bait was laid directly for 30 days. Two weeks later the rat population was census baited using a formulated wheat bait. Further population samples were taken 11, 12 and 22 weeks after the end of the BDH 10131 treatment. The rats caught at each pre- and post-treatment sampling were sexed, weighed and examined for body condition; they were then autopsied to determine their reproductive status.

The average amount of BDH 10131-treated bait consumed ranged between 103 and 400 g/day. The census bait was more readily eaten and during the last 5 days of the 23-day census period 9-10 kg of wheat was taken daily. The capture of sub-adult animals and actively breeding females at each post-treatment sampling showed that the direct BDH 10131 treatment failed to prevent successful reproduction. In contrast, prolonged suppression of reproduction occurred following an earlier treatment on rats that were fed plain "pre-bait" before BDH 10131 was included.

The results of the two different treatments are compared and considered in relation to the application of BDH 10131 and similar oestrogenic steroids against rats.

Sacco, A. G. 1979. Inhibition of fertility in mice by passive immunization with antibodies to isolated zonae pellucidae. *Journal of Reproduction and Fertility* 56:533-537.

CLINIC, FEMALE, IMMUNOCONTRACEPTION, MAMMAL, MOUSE, ZONA PELLUCIDA

"A chromatographically purified preparation of gamma globulin produced against isolated zonae pellucidae of mice was used to immunize mice. A single 2.5 mg dose totally inhibited fertility for a minimum of 11 days." (p. 533)

Sacco, A. G. 1987. Zona pellucida: current status as a candidate antigen for contraceptive vaccine development. *American Journal of Reproductive Immunology and Microbiology* 15:122-130.

IMMUNOCONTRACEPTION, OVERVIEW, ZONA PELLUCIDA**Abstract**

The porcine zona pellucida represents a unique immunocontraceptive target antigen. Initial active immunization studies using crude or partially purified zona components as immunogens resulted in adverse, nonreversible effects on ovarian folliculogenesis, but more recent findings in nonhuman primates indicate that such effects are not as severe and are reversible. Consequently, current efforts are directed toward identifying a zona immunogen that elicits contraceptive antibodies that produce no adverse effects on the ovary. Other studies are in progress to determine optimal injection/immunization format, proper immunogen dosage, and appropriate adjuvant for use in the zona-antizona immunocontraceptive system. Recent data have been very encouraging and emphasize the contraceptive efficacy of this approach to reproduction control.

Sacco, A. G., and C. A. Shivers. 1978. Immunologic inhibition of development. Pages 203-228 in J. C. Daniel, editor. *Methods in Mammalian Reproduction*. Academic Press, New York.

IMMUNOCONTRACEPTION, OVERVIEW

This chapter presents an overview of immunocontraception, focusing on the physiological and endocrinological pathways that make this approach to contraception feasible.

"The inhibition of fertility and development by immunologic means is a broad and continuously expanding area of research. It is beyond the scope of the present chapter to describe in intimate detail the myriad procedures involved for the preparation of different antigens, production and collection of antisera, testing antisera specificity, absorbing antisera, and testing antisera effect both on the target antigen and on fertilization and development. The major purpose of this chapter was to familiarize the reader with the concepts of immunocontraception, and to present a description of the procedures one should take to explore this approach to fertility regulation. The antigens associated with the ovary and zona pellucida were used as the model system.

"For those interested in working in the area of immunocontraception, two topics discussed in this chapter warrant reemphasis due to their critical importance to this approach of reproduction control. These include (1) the absolute necessity for establishing the tissue specificity of the antigen involved by the examination of numerous tissues by several different techniques such as immunodiffusion, immunofluorescence, RIA, etc.; (2) the need to reconfirm *all* findings using purified [IgG or F(ab')₂ fractions] antiserum preparations. By this critical examination of specific antigens that may be present in reproductive tissues, there exists the potential for a new and effective means of fertility regulation."

Sacco, A. G., M. G. Subramanian, and E. C. Yurewicz. 1981. Active immunization of mice with porcine zonae pellucidae: Immune response and effect on fertility. *Journal of Experimental Zoology* 218:405-418.

CLINIC, FEMALE, IMMUNOCONTRACEPTION, MAMMAL, MOUSE, PZP, ZONA PELLUCIDA

Abstract

Female mice were heteroimmunized with varying dosages of intact and heat-solubilized porcine zonae. Mouse immune response was monitored by radioimmunoassay and the effect on mouse fertility of circulating antibodies to pig zonae was observed.

Porcine zonae were a potent heteroimmunogen in the mouse and, for all dosages studied, the immune response was characterized by a rapid rise in titer, culminating in a peak which was followed by a plateau period of extended duration and of significantly high anti-zona activity. Maximum immune response was achieved using a dosage of 4000 heat-solubilized zonae per mouse. Studies involving alteration of immunization regimes, but use of a constant zonae dosage, indicated that maximal response to this antigenic target was obtained using intact zonae applied over a series of injections rather than in a single application.

In vitro studies involving proteolytic lysis of or sperm attachment to mouse zonae pretreated with either antibodies to mouse zonae (homologous system) or pig zonae (heterologous system) demonstrated that zona antibodies are considerably more biologically effective in the homologous system.

Immunization of mice with pig zonae had no significant effect upon altering mouse fertility. The significance of these fertility results is discussed with respect to the information obtained from the in vitro

studies and in respect to the continued consideration of the zona as an immunocontraceptive antigenic target.

Sacco, A. G., E. C. Yurewicz, M. G. Subramanian, and F. J. Demayo. 1981. Zona pellucida composition: species cross reactivity and contraceptive potential of antiserum to a purified pig zona antigen (PPZA). *Biology of Reproduction* 25:997-1008.

CLINIC, IMMUNOCONTRACEPTION, MAMMAL, MONKEY, MOUSE, PIG, PZP, RABBIT, RAT, ZONA PELLUCIDA

Abstract

Antiserum produced against...PPZA, purified pig zona antigen isolated from porcine zonae was examined in respect to its species cross reactivity and contraceptive potential. Antiserum to PPZA produced precipitation layers of varying intensities on the zonae of all species tested. The degree of cross reactivity of anti-PPZA serum for zonae from various species, estimated by precipitation and immunofluorescent titrations, was in the order of human>squirrel monkey>rabbit>rat>mouse. ...Pretreatment of intact zonae from human and squirrel monkey with PPZA antibody resulted in the in vitro inhibition of homologous sperm adherence to such treated zonae, thus demonstrating the contraceptive potential of PPZA antiserum in these two species. Similar treatments using the rabbit and mouse systems did not show any observable inhibition of homologous sperm adherence.

...Collectively...electrophoretic and immunologic studies suggest that the zonae of pig, human, squirrel monkey, and rabbit each contain an acidic macromolecule which possesses antigenic determinants similar to those of PPZA.

Sacco, A. G., M. G. Subramanian, E. C. Yurewicz, F. J. DeMayo, and W. R. Dukelow. 1983. Heteroimmunization of squirrel monkeys (*Saimiri sciureus*) with a purified porcine zonae antigen (PPZA): immune response and biologic activity of antiserum. *Fertility and Sterility* 39:350-358.

CLINIC, IMMUNOCONTRACEPTION, MAMMAL, MONKEY, PZP

Abstract

The potential for utilization as a contraceptive vaccine of...purified porcine zona antigen (PPZA)...was investigated in the squirrel monkey. Immunization resulted in production and maintenance of high antibody titers for at least 1 year. Comparable immune profiles were obtained using either monkey or pig zonae in assay systems, but dose-dependent variations in immune response were not observed. In situ antibody binding to monkey zonae was detected, but significantly fewer ovulated eggs were obtained from immunized monkeys than from controls. Exposure of antibody-pretreated pig, monkey and human zonae to homologous sperm resulted in total inhibition of sperm attachment for the respective species. Thus, the contraceptive potential of PPZA antibodies in these species is demonstrated.

Sacco, A. G., M. G. Subramanian, and E. C. Yurewicz. 1984. Association of sperm receptor activity with purified pig zona antigen (PPZA). *Journal of Reproductive Immunology* 6:89-103.

CLINIC, MALE, PIG, PZP, ZONA PELLUCIDA

Abstract

Three types of data are presented which suggest that sperm receptor activity is associated with a 58,000 Mr acidic glycoprotein (PPZA) isolated from pig zona: (1) boar sperm fail to bind to pig zona pretreated with univalent antibody to PPZA; (2) boar sperm pretreated with PPZA are inhibited in binding to intact pig zona; (3) binding studies utilizing boar sperm and radio labeled PPZA indicate an inhibition of radio labeled PPZA binding to sperm by non-labeled PPZA in a dose-related manner. Collectively these data suggest that, at least in the pig system, the contraceptive action of zona antibodies is accomplished by a specific immunologic reaction between antibody and the zona sperm receptor site rather than to a non-specific blockage of this receptor.

Saini, M. S., and V. R. Parshad. 1988. Laboratory evaluation of a toxicant-sterilant α -chlorohydrin for the control of Indian mole rat *Bandicota bengalensis*. *Annals of Applied Biology* 113:307-312.

ALPHA-CHLOROXYDRIN, CHEMOSTERILANT, CLINIC, MAMMAL, RAT

"A toxicant-sterilant, α -chlorohydrin, was evaluated against the Indian mole rat *Bandicota bengalensis* and the ship or house rat *Rattus rattus*. It caused 100% mortality of *B. bengalensis* at 100 mg/kg but no mortality was observed in *R. rattus* even at 300 mg/kg. The survivors of *B. bengalensis*, which received 60-90 mg/kg of α -chlorohydrin by oral intubation, showed dose-dependent decreases in testicular weight, and cauda epididymal sperm concentration, live sperm count and sperm motility. The values of these parameters indicated that mole rats receiving the above doses would be sterile. In contrast, these changes were observed in *R. rattus* only at doses of 200 and 300 mg/kg. The toxic and antifertility effects of α -chlorohydrin observed on *B. bengalensis* suggest that it should be evaluated for the management of this species under field conditions." (p. 307)

Samojlik, E., and M. C. Chang. 1970. Antifertility activity of 3-chloro-1, 2-propanediol (U-5897) on male rats. *Biology of Reproduction* 2:299-304.

ALPHA-CHLOROXYDRIN, CHEMOSTERILANT, CLINIC, MALE, MAMMAL, RAT, U-5897

Abstract

Sterility in male rats was shown after subcutaneous injections of 3-chloro-1, 2-propanediol...at a dose of 15 mg/kg daily for 6 days and a dose of 40 mg/kg for 3 days. Recovery of fertility, however, was observed 18 days after treatment for 30 days at a dose of 15 mg/kg. The higher dose daily for 10 and 15 days caused a marked reduction of the period of motility of epididymal spermatozoa *in vitro*, a marked drop of sperm count in the cauda epididymis, and a significant decrease of oxygen uptake by sperm. The same dose daily for 7 or 20 days induced morphological changes of the caput and cauda epididymis, the testis, and their blood vessels; recovery of fertility was not observed 140 days after the cessation of the 20 day treatment. ...The pituitary gonadotrophins showed no visible change. The treated males exhibited normal sex drive and produced vaginal plugs after mating.

Sanborn, W. A., R. H. Schmidt, and H. C. Freeman. 1994. Policy considerations for contraception in wildlife management. *Proceedings of the Vertebrate Pest Conference* 16:311-316.

HUMAN DIMENSIONS, POLICY, REGULATION

Abstract

Managing wildlife populations by manipulating their birth rates is a promising technology. However, the use of contraceptive technologies will involve the development of new wildlife management policies. We designed and implemented a survey that was intended to gather information on the range of perspectives of concerned publics on contraceptive use in wildlife management. There appears to be considerable confusion and mistrust regarding the application and appropriateness of this new technology. We recommend that promoters of contraception use in wildlife management be careful to explain what this new technology can and cannot do in order to avoid the pitfalls associated with trying to deliver false promises.

Schafer, E. W., R. B. Brunton, and N. F. Lockyer. 1976. Evaluation of 45 chemicals as chemosterilants in adult male quail (*Coturnix coturnix*). *Journal of Reproduction and Fertility* 48:371-375.

AVES, CHEMOSTERILANT, CLINIC, MALE, QUAIL

Of the 45 compounds tested, 13 resulted in significantly reduced fertility or sterility. Further tests (on males with regressed testes) of the 12 compounds that reduced fertility in breeding males identified 3 compounds that reduced fertility and 5 that caused sterility. The most active of these, thiotepa, was chosen for additional tests with starlings (*Sturnus vulgaris*).

Schafer, E. W., R. B. Brunton, E. C. Shafer, and G. Chavez. 1982. Effects of 77 chemicals on reproduction in male and female *Coturnix* Quail. *Ecotoxicology and Environmental Safety* 6:149-156.

AVES, CHEMOSTERILANT, CLINIC, FEMALE, MALE, QUAIL

Seventy-one chemicals were administered as single oral doses at about 50% of the estimated LD50 to adult male coturnix quail (*Coturnix coturnix*). None reduced the fertility of eggs produced by female mates by more than 50%. Of six additional chemicals similarly administered to female quail at 24 to 56% of the estimated LD50 only one, P,P-bis(1-aziridinyl)-N-phenylphosphinic amide, reduced expected egg fertility by more than 50%.

Schmidt, R. 1993. Emerging technologies: contraception and wildlife management. *ADC* 1(6):8-9.

HUMAN DIMENSIONS, IMMUNOCONTRACEPTION, OVERVIEW

A brief overview written for ADC practitioners. The article explains some of the motivating forces behind contraceptive research, including humane considerations, "location, season, timing, cost, endangered or threatened species considerations, or regulatory concerns..." that may prohibit the use of conventional population control methods. The article also discusses "the form and mode of action of contraceptive agents, the method of delivery, and the social and policy considerations relating to contraception in wildlife management."

Schortemeyer, J. L., and S. L. Beckwirth. 1970. Chemical control of pigeon reproduction. *Transactions of the North American Wildlife and Natural Resources Conference* 35:47-55.

AVES, CHEMOSTERILANT, FIELD, ORNITROL, PIGEON

Results of this study were somewhat ambiguous, and it is difficult to attribute all of the population decline to Ornitrol; the untreated control site experienced a 23% reduction in population.

"Although the St. Augustine (control) flock underwent a 34 percent reduction in population, the 57 percent reduction at both Ocala and the Jacksonville Ice Plant are sufficiently greater to attribute a large part of the difference to the effect of Ornitrol. Furthermore, it seems reasonable to assume that the downward trend in St. Augustine is an annual fluctuation, and that by June, 1970, the control population will be back up to its original level of May, 1969. The major problem to date seems to be the fact that the first treatment was not given until April while it should have been administered as early as December, 1968, or January, 1969." (p.54)

Seal, U. S. 1991. Fertility control as a tool for regulating captive and free-ranging wildlife populations. *Journal of Zoo and Wildlife Medicine* 22:1-5.

OVERVIEW, POLICY

An overview of some of the concerns and constraints surrounding the application of contraceptive techniques on free-ranging animals.

"Fertility control holds promise as one tool for the demographic and genetic management of captive and wild populations. Applications to wild populations need methods that (1) are reversible, (2) can be administered in a single application, (3) can be administered remotely with a minimum of trauma to the animal, (4) should have a controllable period of effectiveness extending up to the life expectancy for individuals of that species, (5) should either not interfere with pregnancy and lactation or be an effective pregnancy termination agent, (6) should not adversely affect social behavior, and (7) should have an acceptable incidence of morbidity. There should be no persistent contraceptive effect on a predator eating the treated animal. The cost of the agent, its vehicle, and its administration should compare favorably with cost of alternate methods of control. Fertility control of selected wild populations permits full use of the genetic stock, allows flexible response to changing population numbers, and can be more effective than culling, which can disrupt social structure, usually generating a population growth response with rapid return to original numbers." (p. 5)

Seal, U. S., R. Barton, L. Mather, K. Oberding, E. D. Plotka, and C. W. Gray. 1976. Hormonal contraception in captive female lions (*Panthera leo*). *Journal of Zoo Animal Medicine* 7:1-17.

CAT, CLINIC, LION, MAMMAL, MEDROXYPROGESTERONE ACETATE, MELENGESTROL ACETATE

Two avenues of contraception were assessed in producing infertility in captive lions. The purpose of the effort was to limit reproduction in captive (zoo) populations. Injectable medroxyprogesterone acetate (MPA), and melengestrol acetate and MPA implants were used. Although both techniques demonstrated reversibility, the authors preferred the implants as being easier to administer.

The authors monitored the subjects closely for signs of side effects reported for these compounds. The highly controlled environment allowed the researchers to mitigate these effects by changing diet or switching to a different contraceptive compound. "The possible appearance of these complications must be considered at each examination as treatment of the animals is continued."

The efficacy of these contraceptive compounds had, as of the writing of this paper, "been evaluated on 64 animals for 787 animal months and 98 animal breeding seasons," with no pregnancies reported. However,

additional questions remain unanswered, including the minimum effective dose (and frequency of dosage) for these compounds.

Setty, B. S., A. B. Kar, S. K. Roy, and S. R. Chaudhury. 1970. Studies with sub-toxic doses of α -chlorohydrin in the male monkey (*Macaca mulatta*). *Contraception* 1:279-289.

ALPHA-CHLOROHYDRIN, CHEMOSTERILANT, CLINIC, MALE, MONKEY

Abstract

A sub-toxic dose of α -chlorohydrin...has been investigated in male rhesus monkeys. At the dosages used the compound has no effect on the genital organs and the pituitary. Extragonadal sperm number, morphology and motility also remain unaltered. There is a pronounced increase in Zn65 uptake and oxygen consumption by vasal spermatozoa. If the dosages used elicit an antifertility effect, the compound may do so by acting directly on spermatozoa and renders them sterile by damaging their membrane.

Short, R. V. 1992. Elephants and birth control. *New Scientist*(1 August):21-23.

ELEPHANT, FEMALE, MAMMAL, OVERVIEW, RU486

An article based on research presented at a symposium held in Kenya in May of 1992. The article summarizes the complexities associated with contracepting elephants, indicating problems associated with various proposed approaches. The article also explains some ecological and human dimension motivations behind the drive to identify an elephant contraceptive.

The author suggests two avenues which may prove useful. One is based on the abortifacient RU486, which, when administered correctly could be effective without disrupting elephant behavior or human sensitivities. In the long term, researchers will attempt to develop anti-zona pellucida vaccine for elephants. The relatively long gestation period and slow reproductive rate of elephants will require a long acting, slow release encapsulated anti-ZP vaccine.

Simmons, J. G., and C. E. Hamner. 1973. Inhibition of estrus in the dog with testosterone implants. *American Journal of Veterinary Research* 34:1409-1419.

DOG, FEMALE, IMPLANT, MAMMAL

"One possible method to obtain reversible long-term contraception in the bitch is the subcutaneous implantation of testosterone in capsules fabricated from silicone rubber and calculated to release different daily doses of testosterone.

"Thirteen bitches were kept in anestrus for 420 to 840 days. In 7 bitches, the testosterone-containing implants were removed at the end of 420 to 480 days and the bitches resumed a normal estrous cycle. Six bitches were bred, and they whelped and weaned normal pups.

"A predictable, effective estrus-inhibiting dose of testosterone was 759 μ g/4.5 kg. of body weight per day.

"Androstenedione, at the dose levels used, was ineffective for inhibiting estrus in the bitch."

Sokolowski, J. H., and F. VanRavenswaay. 1976. Effects of melengestrol acetate on reproduction in the beagle bitch. *American Journal of Veterinary Research* 37:943-945.

DOG, FEMALE, MAMMAL, MELENGESTROL ACETATE

"Melengestrol acetate orally given to bitches at dose levels of 200 µg or more each day effectively inhibited oestrous activity for a treatment period of 243 days and a lapsed period of more than 400 days from the preceding estrous period. The 100-µg dose of the drug, given orally once a day, starting immediately after the bitches were bred or late in gestation, had no effect on conception, pregnancy, or parturition. Gestation was prolonged in bitches given melengestrol acetate starting 1 day after breeding. Drug-polymeric implants blocked reproductive activity, and the progestogen induced cystic endometrial hyperplasia."

Sokolowski, J. H., and R. G. Zimbelan. 1976. Evaluation of selected compounds for estrus control in the bitch. *American Journal of Veterinary Research* 37:939-941.

DOG, FEMALE, MAMMAL, MIBOLERONE, STEROID

"Steroidal and nonsteroidal compounds were evaluated for estrus inhibition in the bitch. Mibolerone, an anabolic steroid, was effective and appeared to be safe. The remaining compounds lacked efficacy or had extensive side effects, or both."

Stellflug, J. N., N. L. Gates, and R. G. Sasser. 1978. Reproductive inhibitors for coyote population control: developments and current status. *Proceedings of the Vertebrate Pest Conference* 8:185-189.

CLINIC, COYOTE, FEMALE, FIELD, MALE, MAMMAL, OVERVIEW

Abstract

...This review deals with a class of possible alternatives for population control (reproductive inhibitors) and the conditions associated with selection and application of reproductive inhibitors to the target species.

Stellflug, J. N., C. W. Leathers, and J. S. Green. 1984. Antifertility effect of Busulfan and DL-6-(N-2-pipecolinomethyl)-5-hydroxy-indane maleate (PMHI) in coyotes (*Canis latrans*). *Theriogenology* 22:533-543.

BUSULFAN, CLINIC, COYOTE, FEMALE, MALE, MAMMAL, PMHI

Abstract

Antifertility effects of busulfan were evaluated using adult coyotes. In addition, antifertility effects of PMHI were evaluated in adult males. Adult males and females were allotted randomly to the following treatments: (1) untreated control, (2) a single oral dose of ...busulfan... or (3) two oral doses of 3 mg busulfan/kg body weight (BW) given nine days apart. The untreated males were used as controls in both experiments. Additional male coyotes were allotted randomly to PMHI treatments as follows: (1) a single oral dose of 2 mg PMHI/kg BW, or (2) two oral doses of 1 mg PMHI/kg BW given seven days apart. Blood samples were taken from females and serum analyzed for progesterone by radioimmunoassay. ...For females developing corpus luteum (CL), the maximum peak progesterone concentration for those given two doses of busulfan was less ($P < 0.05$) than that for untreated controls and single-dose females (16.9, 22.4 and 26.3 mg/ml, respectively). The double treatment of busulfan prevented more females (4 of 10) from developing CL ($P < 0.08$) than controls (0 of 7) or single-dose females (1 of 9). None of the busulfan-treated male coyotes had histologic evidence of spermatogenesis 60 days after the onset of treatment. The oral dose or doses of PMHI did not result in complete degeneration of seminiferous tubules. Busulfan given