

#### Canine sexing agent - MALE

### PRODUCT: PUPPLUS-M

#### INDICATIONS

For producing more male pups in dogs.

#### DOSAGE

One insemination unit of *extended* fresh, chilled, or frozen semen per vial. Recommended extension volume = 2.5 ml/vial. Recommended titer: 100 x 10<sup>6</sup> live sperm/ml.

#### NOTICE

PUPPLUS-M is a non-prescription biopharmaceutical agent. Federal law *does not* require that this product be used by or on the order of a licensed veterinarian. This is not a restricted drug.

#### DESCRIPTION

PUPPLUS-M is spermagenic agent for sexing a single dose of fresh, chilled, or frozen dog semen. Packaged in kit form, each dose is sealed in a vial to maintain potency during storage. The agent is activated by adding extended semen directly to the PUPPLUS-M vial. After a short incubation, the sexed semen is ready for insemination.

#### MODE OF ACTION

PUPPLUS-M works by enhancing the fertility of the Y-chromosome bearing (male) sperm while reducing the motility of the X-chromosome bearing (female) sperm. When inseminated near the time of ovulation, the sperm sort in the reproductive tract of the bitch and more ova are fertilized by the Y-chromosome bearing (male) sperm. The MALE SEX RATIO is increased by 20-30% and fertility is boosted 5-20%.

**DETERMINE SEMEN PARAMETERS:** It is recommended to determine semen parameters prior to mixing with PUPPLUS-M or in a parallel sample.

#### KIT INSTRUCTIONS FOR CHILLED OR FROZEN SEMEN

- When sexing chilled semen, warm semen to 95-98.6°F (35-37°C) in water bath.
- When sexing frozen semen, thaw as usual and pool samples into a single sterile tube. Warm to 95-98.6°F (35-37°C) in a water bath.
- Determine semen parameters and adjust if necessary. Note: Be sure to evaluate sperm prior to mixing with PUPPLUS-M.
- Warm PUPPLUS-M vial to 95-98.6°F (35-37°C) for several minutes to prevent cold shock to semen.
- Introduce 2-3 ml of thawed dog semen into the PUPPLUS-M vial.
- Reseal vial and gently mix semen with contents of vial.
- Return PUPPLUS-M + semen to water bath.
- IMPORTANT: Incubate the semen for 20-30 minutes at 95-98.6°F (35-37°C) prior to insemination.
- Load into insemination pipette and inseminate.

### KIT INSTRUCTIONS FOR FRESH SEMEN

- Dilute freshly collected semen with a suitable canine extender to 100 x 10<sup>6</sup> live sperm/ml and equilibrate to 95-98.6°F (35-37°C) for 10 minutes.
- Warm PUPPLUS-M vial to 95-98.6°F (35-37°C) in water bath to prevent cold shock.
- Introduce 2-3 ml of extended dog semen into the PUPPLUS-M vial. Reseal the PUPPLUS-M vial and gently mix.
- Immediately incubate the PUPPLUS-M and semen in a water bath or incubator for 20-30 minutes at 95-98.6°F (35-37°C).
- Load insemination pipette with sexed semen and inseminate.

Note: During incubation, keep vial sealed to protect from light, air and water.

## **GOOD BREEDING PRACTICES**

- Good reproductive management of the bitch is important for best results.
- Good heat detection prior to and during the time of breeding will improve results.
- PUPPLUS-M is a sperm fertility agent. Overall bitch fertility will likely be higher when using PUPPLUS-M. Number of pups may increase.

### **BREEDING PROTOCOL WITH PUPPLUS-M**

Timing of the insemination is important for best results when using PUPPLUS-M.

- Delay insemination until ova are ovulated and fully mature. This will
  increase the number of male pups. When insemination is delayed until
  ova undergo maturation (30 hrs. after ovulation), the male sperm can
  immediately fertilize the mature ovum.
- On the contrary, when sexed sperm are inseminating too early in the heat, the male sperm undergo senescence and will be unable to fertilize the ova.
   Fewer male pups may result.

LH Surge: Inseminate 6 days (144 hours) following the LH surge.

**Progesterone rise:** Inseminate 24-36 hours following a rise in progesterone levels to 32-64 nmol/L. See Table 1.

NOTE: Optimal fertility is anticipated when serum progesterone is between 32-64 nmol/L at the time of breeding. In comparing progesterone results from different sources, be aware that progesterone concentrations may be reported in units of 'ng/ml' or 'nmol/L'. To convert results into similar units, use the following:

- Progesterone in ng/ml x 3.18 = Progesterone in nmol/L
- Progesterone in nmol/L ÷ 3.18 = Progesterone in ng/ml

Table 1

Ranges of Progesterone Results and Their Implication for Breeding
('Days' refers to the number of days after collection of the sample)

Concentration of Progesterone	Estimate for Ovulation (no. of days)	Estimated Time for Breeding (no. of days)
0-2 nmol/L	Baseline concentration, too early to estimate ovulation	Not applicable
3-6 nmol/L	Minimum of 2 days before ovulation is expected. Results of 3-4 nmol/L may persist for a week or longer before increasing	Earliest estimated window for breeding is from 4-6 days, but could be longer
7-12 nmol/L	Minimum of 1 day before ovulation is expected	Estimated window for breeding is from 3-5 days, but could be longer
13-18 nmol/L	Ovulation is impending or has just occurred	Estimated window for breeding is 2-4 days
19-31 nmol/L	Ovulation recently occurred	Estimated window for breeding is 1-3 days
32-64 nmol/L	Ova have matured, optimal potential for fertility	Estimated window for breeding is 0-2 days
65-90 nmol/L	Ova have matured but aging, decreased potential for fertility	Breed at once (0-1 days)
>90 nmol/L	Too late or very reduced potential for fertility	

WEBCD.ENDO.REF Progesterone Guidelines

## STORAGE CONDITIONS

Store in freezer (-20°C). Avoid moisture and strong sunlight.

# HOW SUPPLIED

PUPPLUS-M is lyophilized in the following package sizes: 2.5 mL vials.

## WARNINGS KEEP OUT OF REACH OF CHILDREN.

Manufactured by:



EMLAB GENETICS, LLC Arcola, IL 61910 USA

For more information, please call 708-442-3964 or log on to www.emlabgenetics.com

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