**A Four-week Repeated Oral Dose Toxicity Study of PROJECT 2 in Dogs**

1. **SUMMARY AND CONCLUSION**

The oral toxicity of PROJECT 2 suspended in 0.5% methylcellulose solution (0.5% MC solution) was investigated in beagle dogs (3/sex/group) administered once daily for 4 weeks at 30, 100, or 300 mg/kg. The control animals received 0.5% MC solution, the vehicle for the test article. The plasma PROJECT 2 concentration was also measured to evaluate systemic exposure.

During the treatment period, general signs were observed, and body weight and food consumption were measured. The animals underwent hematological examination, blood chemistry testing, urinalysis, ophthalmologic examination, electrocardiography, and toxicokinetic analysis. In addition, gross pathological examination, organ weight measurement, and histopathological examination were conducted at the completion of the treatment period.

Clinical signs included grayish white soft feces observed in one male dosed with 100 mg/kg for 1 day, and grayish white feces (including grayish white watery feces in one female for 1 day) that were noted for 1 to 10 days in all animals dosed with 300 mg/kg . The grayish white colored feces were considered to be due to unabsorbed test article. For the following items, no toxic changes occurred up to 300 mg/kg: body weight, food consumption, hematological examination, blood chemistry testing, urinalysis, ophthalmological examination, electrocardiography, gross pathological examination, organ weight, and histopathological examination.

Plasma PROJECT 2 concentration data were as follows: On Day 1, the Cmax values for the males and females were, respectively, 3.189 and 1.552 ng/mL in the 30 mg/kg group; 2.139 and 3.297 ng/mL in the 100 mg/kg group; and 4.756 and 4.315 ng/mL in the 300 mg/kg group. The AUC0-24h values for the males and females were, respectively, 16.100 and 4.310 ng∙h/mL in the 30 mg/kg group; 9.656 and 9.726 ng∙h/mL in the 100 mg/kg group; and 15.852 and 13.920 ng∙h/mL in the 300 mg/kg group. The Tmax values were between 0.5 and 8.3 h for both the males and females. During Week 4, the Cmax values for the males and females in the 30, 100, and 300 mg/kg groups were 4.790 and 2.526 ng/mL, 3.451 and 8.406 ng/mL, and 3.526 and 8.342 ng/mL, respectively. The AUC0-24h values for the males and females in the 30, 100, and 300 mg/kg groups were 16.875 and 9.991 ng∙h/mL, 24.043 and 35.602 ng∙h/mL, 27.219 and 49.570 ng∙h/mL, respectively. The Tmax values were between 0.5 and 1.7 h for both the males and females. On Day 1 and during Week 4, there were no clear differences in the Cmax or AUC0-24h values among the doses. In addition, comparison of the Cmax and AUC0-24h values between Day 1 and Week 4 indicated no clear difference after repeated administration.

To summarize, the no-observed-adverse-effect-level for this study was judged to be 300 mg/kg/day.