Effects of sulfamethoxazole-trimethoprim on thyroid function in dogs

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Objective—To evaluate effects of trimethoprim-sulfamethoxazole (T/SMX) on thyroid function in dogs.

Animals—6 healthy euthyroid dogs.

Procedure—Dogs were administered T/SMX (14.1 to 16 mg/kg, PO, q 12 h) for 3 weeks. Blood was collected weekly for 6 weeks for determination of total thyroxine (TT_4), free thyroxine (fT_4), and canine thyroid-stimulating hormone (cTSH) concentrations. Schirmer tear tests were performed weekly. Blood was collected for CBC prior to antimicrobial treatment and at 3 and 6 weeks.

Results—5 dogs had serum TT_4 concentrations equal to or less than the lower reference limit, and 4 dogs had serum fT_4 less than the lower reference limit after 3 weeks of T/SMX administration; cTSH concentrations were greater than the upper reference limit in 4 dogs. All dogs had TT_4 and fT_4 concentrations greater than the lower reference limit after T/SMX administration was discontinued for 1 week, and cTSH concentrations were less than reference range after T/SMX administration was discontinued for 2 weeks. Two dogs developed decreased tear production, which returned to normal after discontinuing administration.

Conclusions and Clinical Relevance—Results suggest that administration of T/SMX at a dosage of 14.1 to 16 mg/kg, PO, every 12 hours for 3 weeks caused decreased TT_4 and fT_4 concentrations and increased cTSH concentration, conditions that would be compatible with a diagnosis of hypothyroidism. Therefore, dogs should not have thyroid function evaluated while receiving this dosage of T/SMX for > 2 weeks. These results are in contrast to those of a previous study of trimethoprim-sulfadiazine. (*Am J Vet Res* 2005;66:256–259)

Sulfonamides in combination with trimethoprim are frequently used in veterinary medicine to treat bacterial infections. **Trimethoprim-sulfadiazine (T/SDZ)** is available as a veterinary-approved product.[a] **Trimethoprim-sulfamethoxazole (T/SMX)** is a human product that exists in a generic formulation. These products are assumed to be equivalent with regard to their dose, efficacy, and adverse effects.[1,2] Trimethoprim-sulfamethoxazole is more frequently used because of its decreased cost and the occasional limited availability of the veterinary product. The suggested dosage for these products ranges from 15 to 30 mg/kg every 12 hours for most infections, and as much as 60 to 90 mg/kg every 12 hours for infections with *Nocardia* spp.[1]

There has been much interest with regard to the effect of trimethoprim-sulfonamide antimicrobials on thyroid function in dogs.[3–7] Dogs are often suspected of having hypothyroidism when they have recurrent bacterial skin infections. Therefore, it is not uncommon for such dogs to be receiving an antimicrobial at the time of thyroid evaluation. Both T/SDZ and T/SMX have been implicated in causing decreased thyroid function in dogs.[4–7] On the basis of the current literature, the effect on thyroid function in dogs appears to be dose related. Trimethoprim-sulfadiazine resulted in clinical hypothyroidism in 2 dogs when administered at approximately 24 mg/kg every 12 hours for 40 and 126 days, respectively,[5,6] and T/SMX altered thyroid function when administered at 30 mg/kg every 12 hours for 42 days.[4] In a recent publication, it was reported that T/SMX, when administered at 26.5 to 31.3 mg/kg every 12 hours, caused decreased **thyroxine (T_4)** concentration after only 1 week of administration.[7] In contrast, when T/SDZ was administered at 15 mg/kg, PO, every 12 hours for 4 weeks, there was no effect on thyroid function.[3] On the basis of these studies, it was assumed that thyroid function could be accurately evaluated in a dog receiving a trimethoprim-sulfonamide combination antimicrobial if administered at 15 mg/kg every 12 hours.

On the basis of clinical observation, we believe that T/SMX administered at 15 mg/kg, PO, every 12 hours alters thyroid function similar to that described when administered at 30 mg/kg, PO, every 12 hours. The purpose of the study reported here was to evaluate the effect of T/SMX on thyroid function in dogs when administered at 15 mg/kg, PO, every 12 hours.

Materials and Methods

Dogs—Six healthy privately owned adult dogs were entered into the study. All dogs were enrolled with the informed consent of their owners. The study was approved by the Institutional Animal Care and Use Committee of the University of Tennesee. Results of CBC and Schirmer tear test and concentrations of **total T_4 (TT_4)** and **canine thyroid-stimulating hormone (cTSH)** were within reference ranges prior to enrollment in the study.

Treatment protocol—Trimethoprim-sulfamethoxazole (14.1 to 16 mg/kg [mean, 14.9 mg/kg], PO, q 12 h) was administered for 3 weeks. Dogs were examined weekly, at which time blood samples were collected for determination of TT_4, **free T_4** (fT_4; determined by use of dialysis), and cTSH concentrations; the Schirmer tear test was also performed. Dogs were examined weekly for an additional 3 weeks after discontinuation of administration of T/SMX, at which time blood samples were collected for thyroid evaluation as

Received May 19, 2004.
Accepted June 11, 2004.
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Table 1—Mean ± SD serum total thyroxine (TT_4) concentration, free thyroxine (fT_4) concentration, and canine thyroid-stimulating hormone (cTSH) concentration in 6 healthy euthyroid dogs administered trimethoprim-sulfamethoxazole for 3 weeks.

Variable	Baseline	Week 1	Week 2	Week 3*	Week 4	Week 5	Week 6	Reference range
TT_4 (nmol/L)	41.83 ± 9.91	30.83‡ ± 11.5	27.67 ± 7.26	13.83† ± 6.08	36.33‡ ± 5.47	38.17‡ ± 8.33	48.67‡ ± 16.68	15–67
fT_4 (pmol/L)	19.17 ± 7.36	12.5 ± 7.09	11.33 ± 4.46	4.5† ± 3.27	14.17 ± 3.76	15.5‡ ± 4.59	20.5‡ ± 10.45	6–42
cTSH (ng/mL)	0.17 ± 0.06	0.18‡ ± 0.06	0.43 ± 0.3	1.08† ± 0.74	0.67† ± 0.43	0.31 ± 0.1	0.23‡ ± 0.1	0–0.68

*Administration discontinued. †Significantly ($P < 0.05$) different from baseline value. ‡Significantly ($P < 0.05$) different from week 3 value.

before. The Schirmer tear test was performed only in dogs in which values had decreased while receiving the antimicrobial. Blood was collected for CBC at 3 and 6 weeks.

Specimen collection and storage—Blood samples for CBC and measurement of TT_4, fT_4, and cTSH were obtained by jugular venipuncture. All samples for endocrine analysis were allowed to clot at room temperature (20°C) prior to centrifugation. Serum was stored frozen at –70°C until all samples were collected. Samples were mailed frozen on ice to a commercial laboratory.[b]

Hormone measurement—The TT_4, fT_4, and cTSH concentrations were determined by a commercial laboratory.[b] Total T_4 was measured by use of a commercially available solid-phase radioimmunoassay kit[c] that has been validated in that laboratory for canine serum.[8] Free T_4 was measured by use of a commercially available kit[d] that has been validated in that laboratory for canine serum.[9] Canine TSH was measured by use of a commercially available immunoradiometric assay[e] that has been validated in that laboratory for canine serum.[8]

Statistical analyses—Repeated-measures ANOVA was used to analyze data for serum TT_4, fT_4, and cTSH concentrations, and WBC and neutrophil counts, by use of a statistical software package.[f] The Kolmogorov-Smirnov test was used to test for normally distributed data. Only fT_4 concentration and neutrophil counts failed normality testing. Friedman repeated-measures ANOVA on ranks was used for data that were not normally distributed. Values were considered significant at $P < 0.05$. Data are reported as mean ± SD values.

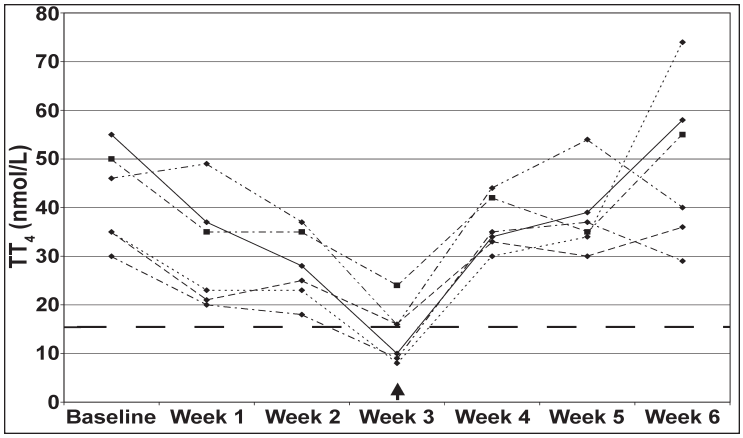

Figure 1—Serum total thyroxine (TT_4) concentrations in 6 healthy euthyroid dogs administered trimethoprim-sulfamethoxazole at a dosage of 14.1 to 14.9 mg/kg, PO, every 12 hours for 3 weeks. Arrow indicates when administration of trimethoprim-sulfamethoxazole was discontinued. Dashed horizontal line denotes lower reference limit.

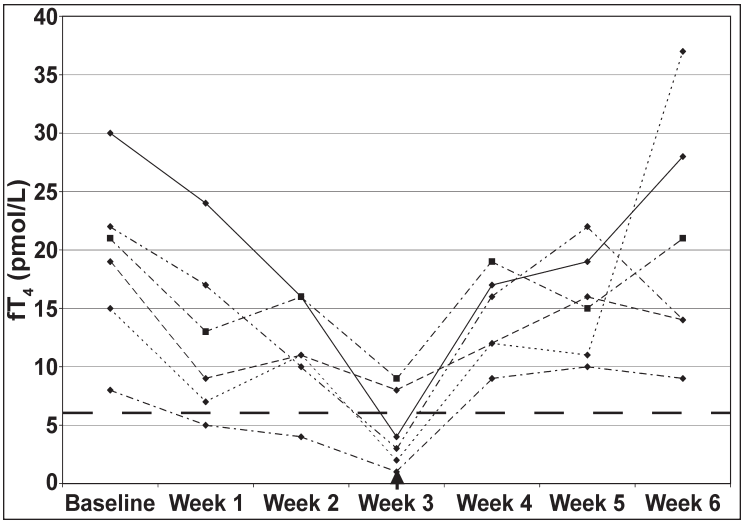

Figure 2—Serum free thyroxine (fT_4) concentrations determined by use of dialysis in 6 healthy euthyroid dogs administered trimethoprim-sulfamethoxazole for 3 weeks. Arrow indicates when administration of trimethoprim-sulfamethoxazole was discontinued. Dashed horizontal line denotes lower reference limit.

Results

Three female (2 spayed and 1 sexually intact) and 3 male dogs (2 castrated and 1 sexually intact) were included in the study (a German Shepherd Dog, a Rottweiler, 2 Brittany Spaniels, and 2 mixed-breed dogs). Dogs ranged from 3 to 7.5 years of age (median, 4.5 years). Body weight ranged from 16.1 to 52.7 kg.

Mean serum TT_4 concentration was significantly ($P < 0.001$) decreased at week 3, compared with pretreatment concentrations (**Table 1**). Serum TT_4 concentrations in 3 dogs were less than the lower reference limit and in 2 dogs were slightly greater than the lower reference limit after 3 weeks of T/SMX administration (**Figure 1**). Mean serum TT_4 concentrations were significantly ($P < 0.001$) greater at weeks 4, 5, and 6 (after discontinuing T/SMX administration), compared with week 3, and were not significantly different from baseline TT_4 concentrations. All 6 dogs had TT_4 concentrations greater than the lower reference limit 1 week after T/SMX administration was discontinued.

Mean serum fT_4 concentration was significantly ($P = 0.003$) decreased at week 3, compared with pretreatment concentrations (Table 1). In 4 dogs, serum fT_4

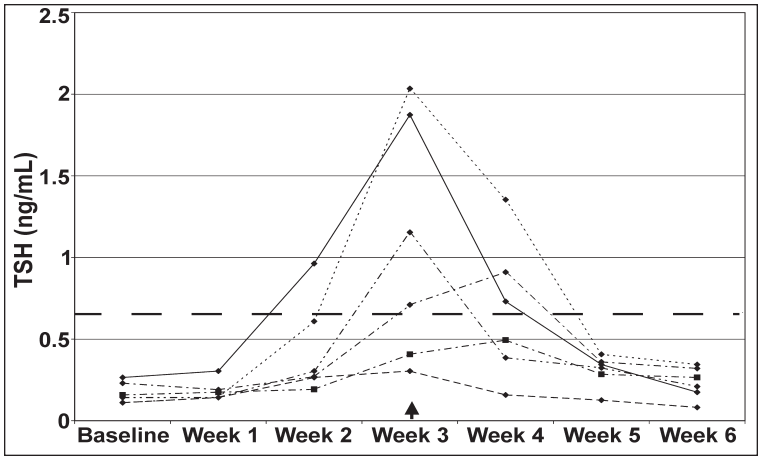

Figure 3—Serum thyroid-stimulating hormone (TSH) concentrations in 6 healthy euthyroid dogs administered trimethoprim-sulfamethoxazole for 3 weeks. Arrow indicates when administration of trimethoprim-sulfamethoxazole was discontinued. Dashed horizontal line denotes upper reference limit.

concentration was less than the lower reference limit after 3 weeks of T/SMX administration **(Figure 2)**. Mean serum fT_4 concentrations were significantly greater at weeks 5 ($P = 0.031$) and 6 ($P = 0.017$), compared with week 3, and were not significantly different from baseline fT_4 concentrations. All 6 dogs had fT_4 concentrations greater than the lower reference limit 1 week after T/SMX administration was discontinued.

Mean serum cTSH concentrations were significantly increased at weeks 3 ($P = 0.018$) and 4 ($P = 0.023$), compared with pretreatment concentrations (Table 1). In 4 dogs, serum cTSH was greater than the upper reference limit after 3 weeks of T/SMX administration **(Figure 3)**. Mean serum cTSH concentration was significantly lower ($P = 0.023$) at week 6, compared with week 3, and was not significantly different from baseline cTSH concentration. Three dogs had cTSH concentrations greater than the upper reference limit at week 4, 1 week after T/SMX administration was discontinued. By week 5, all 6 dogs had cTSH concentrations within the reference range.

The WBC count (8,950 ± 2,569 cells/µL) and neutrophil count (5,348 ± 1,929 cells/µL) were lower at week 3, compared with baseline counts (9,767 ± 2,150 cells/µL and 6,260 ± 1,540 cells/µL, respectively), but not significantly. This was observed in 5 of 6 dogs; however, only 1 dog had a neutrophil count < 2,000 cells/µL (1,820 cells/µL) at week 3. The WBC and neutrophil counts were significantly greater at week 6 (10,916 ± 1,974 [$P = 0.055$] cells/µL and 7,243 ± 1,227 [$P = 0.042$] cells/µL, respectively), compared with week 3.

Tear production decreased to 5 mm/min in the right eye and 10 mm/min in the left eye in 1 dog at week 3. This dog had developed mild mucoid ocular discharge just prior to the week 3 evaluation. Tear production had increased to 15 mm/min in the right eye and 13 mm/min in the left eye 1 week after T/SMX administration was discontinued and was normal by week 5 (2 weeks after T/SMX administration was discontinued). Tear production in another dog decreased to 15 mm/min in the right eye (borderline normal) at week 3. At week 5, tear production was normal in the right eye but had decreased in the left eye (19 and 10 mm/min, respectively). This dog had no clinical evidence of decreased tear production. Tear production returned to normal in the left eye at week 6.

Discussion

Trimethoprim-sulfonamide antimicrobials are known to affect thyroid function in species that are sensitive to its effects, including rats[10,11] and dogs.[4–7] Sulfonamides alter thyroid function by blocking the organic binding of iodine in the thyroid gland by inhibiting thyroid peroxidase.[12,13] This has a direct effect on thyroid production of T_4, resulting in decreased concentrations of both TT_4 and fT_4. Results of our study suggest that administration of T/SMX in healthy dogs decreases thyroid function when administered at 14 to 16 mg/kg, PO, every 12 hours. At this dosage, mean TT_4 and fT_4 concentrations were significantly decreased and cTSH was significantly increased after 3 weeks of T/SMX administration. The effects on T_4 concentration at the lower dosage were not as rapid as that seen when T/SMX was administered at 26.5 to 31.3 mg/kg, PO, every 12 hours; 3 of 6 dogs administered that dosage had decreased concentrations of TT_4 after only 1 week.[7] Therefore, when treating diseases that do not require long-term administration (> 2 weeks), T/SMX administration at 15 mg/kg, PO, every 12 hours should have minimal effect on thyroid gland function. However, for conditions such as pyoderma, in which 3 weeks is the minimum course of treatment, one can expect thyroid gland function to be altered, even at the lower dose.

The TT_4 and fT_4 concentrations had returned to reference range 1 week after discontinuation of T/SMX administration. The TSH concentration did not return to within reference range until 2 weeks after discontinuation of administration, at which time all dogs had TSH concentrations within reference range. These results are similar to those reported when T/SMX was administered at the higher dose of 26.5 to 31.3 mg/kg, PO, every 12 hours.[7] The exact withdrawal time of T/SMX required before assessing thyroid gland function via measurement of T_4 and cTSH is variable (2 to 12 weeks), depending on dosage, duration of administration, and individual variation.[4,5,7]

Results of our study are in direct contrast with those of another study,[3] in which T/SDZ was administered at 15 mg/kg, PO, every 12 hours for 28 days and had no effect on TT_4 concentration, fT_4 concentration, or triiodothyronine concentrations in healthy research dogs. With the exception of the use of research dogs housed in indoor runs and the choice of drug in that study,[3] there was little difference between the 2 studies. Trimethoprim in combination with sulfadiazine is available as a veterinary formulation.[a] Trimethoprim in combination with sulfamethoxazole is presumed to be similar with regard to dosage, spectrum of activity, and adverse effects[1,2] and is available as a generic formulation. Both contain a rapidly absorbed and excreted sulfonamide in combination with trimethoprim in a 5:1 ratio.[14] Both are reported to be well absorbed from the gastrointestinal tract; however, specific data with regard to T/SMX in dogs are lacking. In 1 study,[15] variable absorption of T/SDZ after oral administration in

dogs was reported, which could explain the difference in results if T/SMX is more readily absorbed. Other explanations for this discrepancy are lacking.

The WBC and neutrophil counts were not significantly different after 3 weeks of T/SMX administration, compared with baseline values, although 1 dog developed clinically relevant neutropenia. In dogs, effects of a trimethoprim-sulfonamide antimicrobial on neutrophil counts appear to be dose related. In a previous study,[16] 1 dog had decreased WBC count and neutropenia when receiving T/SDZ at 45 mg/kg, PO, every 12 hours for 8 weeks. In another more recent study[7] in which T/SMX was administered at 26.5 to 31.3 mg/kg, PO, every 12 hours for 3 weeks, 4 dogs developed neutropenia while receiving the drug. The trimethoprim component is most likely responsible for the hematologic abnormalities.[17] Trimethoprim interferes with folate metabolism by inhibiting dihydrofolate reductase.[17] Trimethoprim has a much higher affinity for bacterial than for mammalian dihydrofolate reductase; however, interaction with the mammalian enzyme may occur. The most common hematologic manifestations associated with trimethoprim administration in humans include thrombocytopenia, leukopenia, and granulocytopenia.[18,19] In a human study,[19] some of the hematologic abnormalities associated with trimethoprim were reversed with the addition of folinic acid. Transient neutropenias have also been reported in humans receiving trimethoprim-sulfamethoxazole.[20]

To assess tear production, we used the scoring system described by Moore,[21] which defines dogs with Schirmer tear test readings ≥ 15 mm/min as having normal production, dogs with 11 to 14 mm/min as having subclinical keratoconjunctivitis sicca, dogs with 6 to 10 mm/min as having mild to moderate keratoconjunctivitis sicca, and dogs with ≤ 5 mm/min as having severe keratoconjunctivitis sicca. On the basis of this interpretation, 2 dogs in our study developed decreased tear production. In 1 dog (a Brittany), the decreased tear production developed in both eyes after receiving T/SMX for 3 weeks and was accompanied by clinical signs. The tear production returned to normal after T/SMX administration was discontinued for 2 weeks. Another dog (a Labrador Retriever cross) developed decreased tear production without clinical signs. Interestingly, the tear production in that dog did not decrease to < 15 mm/min until 2 weeks after T/SMX administration was discontinued. Tear production returned to normal in that dog 1 week later. Sulfonamides have a direct toxic effect on lacrimal acinar cells and decrease tear production with dosages ranging from 8.6 to 104 mg/kg/d and as early as 7 days after beginning administration of the antimicrobial.[22] The decreased tear production may be transient and fluctuate during administration of the antimicrobial, or tear production may be permanently suppressed, despite discontinuation of administration.[22,23] Our results support previous findings and emphasize the need to monitor tear production while administering trimethoprim-sulfonamides, regardless of dosage or duration of administration.

a. Tribrissen, Schering-Plough Animal Health Corp, Union, NJ.
b. Diagnostic Center for Population and Animal Health, Endocrine Diagnostic Section, College of Veterinary Medicine, Michigan State University, East Lansing, Mich.
c. Clinical Assays Gammacoat M Total T4 125I RIA Kit, DiaSorin Inc, Stillwater, Minn.
d. Free T4 by equilibrium dialysis, Nichols Institute Diagnostics, San Juan Capistrano, Calif.
e. Coat-A-Count canine TSH IRMA, Diagnostic Products Corp, Los Angeles, Calif.
f. SigmaStat 3.0 for Windows, SPSS Inc, Chicago, Ill.

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