

# Degradation of Estrogen Conjugates Using Titanium Dioxide as a Photocatalyst

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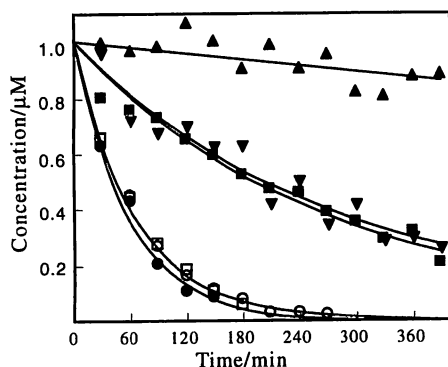


Fig. 2 Photocatalytic degradation of estrogens using photocatalyst under UV irradiation ( $n = 2$ , mean). ●, E<sub>2</sub>; ○, E<sub>1</sub>; ■, E<sub>2</sub>3G; □, E<sub>2</sub>17G; ▼, E<sub>1</sub>G; ▲, E<sub>1</sub>S.

monitored at 280 nm. The flow rate and column temperature were set at 1.0 ml min<sup>-1</sup> and 40°C, respectively. As a mobile phase, methanol-water (7:3, v/v) was used for the determination of unconjugated estrogens [E<sub>2</sub> ( $t_R$  4.3 min) and E<sub>1</sub> ( $t_R$  4.6 min)], and methanol-water containing 5 mM of HCO<sub>2</sub>NH<sub>4</sub> was used for the determination of conjugated estrogens {E<sub>1</sub>S (1:1, v/v;  $t_R$  5.3 min), E<sub>2</sub>G [2:3, v/v;  $t_R$  7.3 min (3G), 7.8 min (17G)] and E<sub>1</sub>G (2:3, v/v;  $t_R$  7.9 min)}.

A VL-4LC black light lamp (365 nm, 4 W) (Vilber Lourmat, Cedex, France) was used as the light source.

#### Photodegradation of estrogens

The ethanol solution of estrogen was diluted with water to 1 µM (the ethanol concentration was less than 0.3%, v/v). Photocatalytic glass beads (ca. 13.6 g) were spread to cover the bottom of a glass petri dish (diameter, 8 cm), and the estrogen solution (1 µM, 15 ml) was placed in the dish. The petri dish was placed in a light shielded box [22.5 (wide) × 13.5 (depth) × 16 cm (height)], and irradiated by a black light lamp 16 cm away for 6.5 h. Five hundred microliters of solution was sampled every 30 min and then subjected to an Oasis HLB cartridge. After washing with water (2 ml), estrogen was eluted with methanol (1.5 ml) and evaporated under a N<sub>2</sub> gas stream. The residue was dissolved in methanol (50 µl) and an aliquot was applied to an HPLC.

#### Recovery of estrogens

The estrogen solutions (1 µM, 500 µl) were subjected to an Oasis HLB cartridge, concentrated and then analyzed by HPLC as described above which were used as the 0 min illuminated samples. The peak areas of the estrogens in these samples were compared with those of the expected amount of authentic estrogens, and the obtained recovery rates (ca. 80%) were used for correction of the concentration in the following experiments.

## Results and Discussion

The time courses of the photocatalytic degradations of the estrogens using the photocatalytic glass beads were monitored by measuring the remaining substrate by UV-HPLC. An aliquot of the reaction mixture was subjected to the solid-phase extraction, concentrated and then analyzed by HPLC, because the UV detector did not have enough sensitivity to detect estrogens without pretreatment.

The results of the photocatalytic degradation are shown in Fig.

2 ( $n = 2$ , mean). E<sub>2</sub>, E<sub>2</sub>17G and E<sub>1</sub>, which have an unconjugated phenolic hydroxy group at the C-3 position, were almost completely degraded after 4 h of UV irradiation by first-order kinetics. Their degradation rate constants were  $1.7 \times 10^{-2} \text{ min}^{-1}$  [coefficient of determination ( $r^2$ ) = 0.984],  $1.5 \times 10^{-2} \text{ min}^{-1}$  ( $r^2$  = 0.991) and  $1.5 \times 10^{-2} \text{ min}^{-1}$  ( $r^2$  = 0.991), respectively. On the other hand, the degradation rates of E<sub>2</sub>3G, E<sub>1</sub>G and E<sub>1</sub>S, which are conjugated at the 3-hydroxy group, were much lower than those of the above estrogens. E<sub>2</sub>3G and E<sub>1</sub>G were degraded slowly by first-order kinetics [ $3.6 \times 10^{-3} \text{ min}^{-1}$  ( $r^2$  = 0.967) and  $3.4 \times 10^{-3} \text{ min}^{-1}$  ( $r^2$  = 0.938), respectively], and ca. 20 and 25% of the substrates remained after a 6.5 h irradiation, respectively. In the case of E<sub>1</sub>S, ca. 90% of the substrate still remained at the end of the irradiation and its degradation order was unclear. E<sub>2</sub> was almost quantitatively recovered from the reaction mixture under the control conditions, such as no UV light or catalyst. These data showed that the adsorption of estrogen on a photocatalyst was negligible and the photocatalyst effectively functioned.

As described above, the hydroxyl radical was generated when TiO<sub>2</sub> was irradiated by UV light in an aqueous solution.<sup>11,12</sup> The oxidation of the phenolic hydroxy moiety of estrogens by the hydroxyl radical was regarded as the initiation of the photocatalytic degradation of the estrogens.<sup>9</sup> The results described in this paper supported the above hypothesis and showed that the 3-hydroxy group of estrogen is important for the photodegradation. The catalytic effect of the other photocatalysts on the degradation of the estrogens conjugated at the 3-hydroxy group and the investigation of the formed degradation products are now in progress in our laboratory.

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#### References

1. Y. Nakamura, K. Onda, C. Takatoh, and A. Miya, *Bunseki Kagaku*, **2003**, *52*, 107.
2. K. Majima, T. Fukui, J. Yuan, G. Wang, and K. Matsumoto, *Anal. Sci.*, **2002**, *18*, 869.
3. G.-G. Ying, R. S. Kookana, and Y.-J. Ru, *Environ. Int.*, **2002**, *28*, 545.
4. F. Tilton, W. H. Benson, and D. Schlenk, *Aquat. Toxicol.*, **2002**, *61*, 211.
5. H. M. Kuch and K. Ballschmiter, *Fresenius J. Anal. Chem.*, **2000**, *366*, 392.
6. K. Wright, D. C. Collins, and J. R. K. Preedy, *Steroids*, **1979**, *34*, 445.
7. K. Shimada, K. Mitamura, and T. Higashi, *J. Chromatogr. A*, **2001**, *935*, 141.
8. T. A. Ternes, H. Andersen, D. Gilberg, and M. Bonerz, *Anal. Chem.*, **2002**, *74*, 3498.
9. Y. Ohko, K. Iuchi, C. Niwa, T. Tatsuma, T. Nakashima, T. Iguchi, Y. Kubota, and A. Fujishima, *Environ. Sci. Technol.*, **2002**, *36*, 4175.
10. H. M. Coleman, B. R. Eggins, J. A. Byrne, F. L. Palmer, and E. King, *Appl. Catal. B: Environ.*, **2000**, *24*, L1.
11. C. Richard and P. Boule, *New J. Chem.*, **1994**, *18*, 547.
12. N. Watanabe, S. Horikoshi, H. Kawabe, Y. Sugie, J. Zhao, and H. Hidaka, *Chemosphere*, **2003**, *52*, 851.