



11175-A FLINTKOTE AVENUE, SAN DIEGO, CA 92121
TELEPHONE (858) 642-1988 • FAX (858) 642-1989
WWW.ATSBIO.COM • ATS@ATSBIO.COM

**Anti-Conjugated 1,3 Butadiene
RABBIT POLYCLONAL**

Catalog Number: AB-T102

ELISA protocol used to test conjugated 1,3-Butadiene:

1. Coating of conjugated 1,3-Butadiene ($10\mu\text{g/ml}$) in maxisorp well plates (Nunc) with a solution of sodium carbonate buffer 0.05M (pH 9.6), during sixteen hours at 4°C .
2. Saturation of well plates with of a solution of PBS (pH 7.3) containing 1g/l of BSA (Acros), 10% of glycerol and 0.5% of Tween (one hour at 37°C).
3. Wash with PBS containing 0.5% of Tween (PBS Tween) (three times).
4. Preabsorbed 1,3-Butadiene antiserum will be diluted (1/5,000-1/10,000) in PBS Tween containing 1g/l BSA, and 10% of glycerol, $200\mu\text{l}$ by well plate (incubating during 2 hours at 37°C).
5. Wash with PBS Tween (three times).
6. $200\mu\text{l}$ of peroxidase-labeled goat anti-rabbit (Biorad) diluted (1/10,000) in a solution of PBS Tween containing 1g/l of BSA, will be applied by well plate (during one hour at 37°C).
7. Well plates will be rinsed with PBS Tween (three times).
8. And finally the peroxidase will be developed by incubating $200\mu\text{l}$ by well plate of a citrate 0.1M/phosphate 0.2M (pH 5) solution containing 0.4% of OPD (Sigma) and 0.03% of hydrogen peroxide (Acros) for ten minutes in the dark, after that, we will stop the reaction by the addition of $50\mu\text{l}$ of 2M HCl.
9. The optical density will be measured at 492nm.