



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

**Anti-Conjugated D-Methionine  
RABBIT POLYCLONAL**

**Catalog Number:** AB-T051

**Example of ELISA protocol used to test conjugated D-Methionine:**

1. Coating of conjugated D-Methionine ( $10\mu\text{g/ml}$ ) in maxisorp well plates (Nunc) with a solution of sodium carbonate buffer  $0.05\text{M}$  (pH 9.6), during sixteen hours at  $4^{\circ}\text{C}$ .
2. Saturation of well plates with of a solution of PBS (pH 7.3) containing  $1\text{g/l}$  of BSA (Acros), 10% of glycerol and 0.5% of Tween (one hour at  $37^{\circ}\text{C}$ ).
3. Wash with PBS containing 0.5% of Tween (PBS Tween) (three times).
4. Preabsorbed D-Methionine antiserum will be diluted ( $1/5,000$ - $1/15,000$ ) in PBS Tween containing  $1\text{g/l}$  BSA,  $1\text{g/l}$  of BSA-G and 10% of glycerol,  $200\mu\text{l}$  by well plate (incubating during 2 hours at  $37^{\circ}\text{C}$ ).
5. Wash with PBS Tween (three times).
6.  $200\mu\text{l}$  of peroxidase-labeled goat anti-rabbit (Jackson) diluted ( $1/10,000$ ) in a solution of PBS Tween containing  $1\text{g/l}$  of BSA, will be applied by well plate (during one hour at  $37^{\circ}\text{C}$ ).
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  $0.1\text{M}$ /phosphate  $0.2\text{M}$  (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  $2\text{M}$  HCl.
9. The optical density will be measured at  $492\text{nm}$ .