

# Cons of a bioassay



**ASSIGN  
BUSTER**

1. Cons of a bioassay- firstly the response to a hormone by a specific tissue may not reflect the 'in vivo' response. For example, a hormone may be modified by another factor within the tissue before it acts on the tissue. Furthermore, the hormone that you are testing i. e the number of contractions in uterine tissue in response to a specific oxytocin concentration, may not be entirely be due to oxytocin i. e. PGF2a is also present in uterine tissue which acts locally to cause contractions also(Callegari et al, 2005). Also, an animal may need to be killed in order to isolate one organ only i. e. the uterus in mice. There is also biological variation between species i. e. human and mouse, and so the physical outcome in response to a particular hormone in mice may not reflect what the outcome is in man.

Pros of a bioassay-allows you to determine whether a hormone is present in a particular tissue and/or plasma. This gives you an idea of how the hormone may act 'in vivo' and how the effects may be modified in a dose response fashion on a particular tissue. The outcomes on hormone responses observed in mice tissue might represent the way the drug works in man(Bulletti, 2005). An unknown concentration of a particular hormone can be devised. You can also use a bioassay to configure the biological response of an exogenous substance, and to make comparisons of the variety of effects of different substances on different tissues.

2. According to Furchgott and Zawadzki (1980) the type of cut of tissue does not matter. As long as the section is carried out carefully so that the endothelial cells remain intact. The myometrium is part of an endothelial layer that contains the receptors for oxytocin. Basically the cut does not matter; however, the receptors need to remain intact.

3. We would automatically think that in the 'absence of any hormones' the baseline activity will be zero. However, this is not the case i. e. if the cotton string was interfered with the baseline activity may be altered. Furthermore, the baseline activity probably is not a reflection of the 'absence of hormones'. Therefore, it is very important to obtain a baseline bioassay measurement, to establish how presence of other factors in the tissue could interfere with subsequent measurements.

4. The greater the amount of oxytocin added to the water bath the greater was the frequency. However, the amplitude did not change. In other words a greater amount of oxytocin added to the water bath-the greater number of contractions within the uterus, although the force of the strength of the contractions was not altered.

5. To quantitate the amount of oxytocin of the unknown (the amount of oxytocin in IU per g in mice pituitary tissue) the area under the curves for each concentration (1IU, 5IU, and 10IU) added to the water bath-the unknown was calculated. The area (uterine work) was figured by printing the curves on graph paper, cutting out the curves and weighing the paper in grams and dividing the weight by the weight of a 1cm<sup>2</sup> piece of paper to give the uterine work (area under curve (cm<sup>2</sup>)). Subsequently, the value of the uterine work for each concentration including the unknown concentration plotted on the graph. Please see graph and calculation on the next page. Calculating the area in this way was a more simple option compared to calculating the work by using the amplitudes (which did not vary considerably) and the frequency, or by counting the number of squares underneath the curves.

6. Please see graph and calculation on following page.

7/8. This assay was not specific. In regards to the uterus in mice other hormones which affect the frequency of contractions are present. For example, PGF<sub>2a</sub> which also acts on the myometrium of the uterus works to increase the amount of contractions. Furthermore, progesterone is responsible for making uterine muscle more contractile, while estrogen and relaxin causes it to relax (Blanks and Shymgol, 2007).

In a pituitary, hormones such as progestins, estrogen's, androgens, which alter uterine contractility, may be found. Additionally, vasopressin is secreted by the posterior pituitary and binds to a receptor very similar to the oxytocin receptor within the uterus (Furchgott and Zawadzki, 1980). At any one time vasopressin may bind to an oxytocin receptor stimulating the same response (Furchgott and Zawadzki, 1980).

Therefore, depending on whether or not and how much of the above hormones were present in the mice uterus and the pituitary- alters the effect of oxytocin acting alone.

Also, other hormones such as prolactin, dopamine, inhibin, LH and FSH are found within the pituitary gland.

9. This bioassay was not good in terms of its specificity as mentioned in questions 7/8. However, it could be made more specific i. e. add antagonists of the hormones present in both the uterine and pituitary tissues to the water bath except for oxytocin, or to isolate the oxytocin from the pituitary tissue

first which is obtained by density gradient centrifugation of isolated pituitary tissue, subsequently adding it to the water bath.

10.

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