

Technical Note, No: 34-0163, v.1.0

## A general instruction for producing additional calibrators

Under certain circumstances it can be beneficial to include additional calibrators when performing analysis with ELISA. This technical note provides information on how to produce additional calibrators from an already existing set of calibrators.

One can choose to add one or more calibrators depending on the number of already existing kit calibrators in the acquired ELISA kit and the need of extra calibrators. To add an additional calibrator from an already existing calibrator set, it is suggested to choose two calibrators next to each other, one with a higher concentration and another one with a lower concentration. The placement of the additional calibrator can for example be between calibrator 1 and 2 or calibrator 4 and 5.

To prepare the new calibrator, pool an equal volume (50:50) of the two existing kit calibrators in a new tube/vial and mix it thoroughly. The new calibrator concentration is calculated by determining the average concentration of the two used kit calibrators (see example in table 1).

Please ensure that you have enough volume of both the old and new calibrators for your planned experiments.

An example where an additional calibrator was added to the original calibrator set of Glucagon ELISA 10-1271-01 is presented below (table 1) together with its calibrator curve (figure 1).

Note: This process is not regularly included in the validation documentation for our products. Thus, it is up to each customer to decide if the new assay needs to be further validated.



Technical Note, No: 34-0163, v.1.0

Table 1: An example of a calibrator set of Glucagon ELISA 10-1271-01 (lot nr: 29870) with a new calibrator 5 generated by pooling an equal volume of the original calibrator 4 and original calibrator 5. Beware that concentration levels of the original kit calibrator of 10-1271-01 may vary between batches with different lot numbers.

Calibrator	Concentration (pM)	Description
Calibrator 1	1.65	Original kit calibrator 1
Calibrator 2	2.87	Original kit calibrator 2
Calibrator 3	8.74	Original kit calibrator 3
Calibrator 4	29.2	Original kit calibrator 4
Calibrator 5	82.6	New calibrator. Generated by pooling the original kit
		calibrator 4 with the original kit calibrator 5 (50:50)
Calibrator 6	136	Original kit calibrator 5

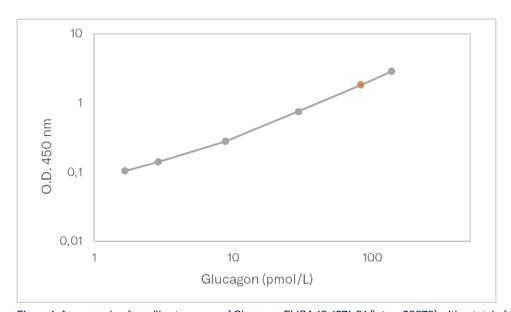


Figure 1: An example of a calibrator curve of Glucagon ELISA 10-1271-01 (lot nr: 29870) with a total of 6 calibrators, whereas the new calibrator 5 (in orange) is generated by pooling the original calibrator 4 and the original calibrator 5.