Intraoperative PTH monitoring: a new approach based on the identification of the “true” time origin of the decay curve

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Abstract. Some published criteria for intraoperative monitoring of PTH serum concentrations may cause misleading results, since the timing of samples measured between the pre-incision and pre-excision phase of surgery is often unrecorded. In our opinion this information is critical, as the time of an intermediate sample during surgical manipulation may represent the “true” beginning of the PTH decay. We modified the usual criterion of monitoring (cut-off at 10 minutes after clamping) proposing a further check at manipulation in case the primary check at clamping produces an apparently negative result. On the basis of a mathematical model, false negative curves were simulated by means of a time shift. This shift was assumed to be the interval between manipulation and clamping. Analysing the decay curve, we used the 50% cut-off at 10 minutes after the supposed “true” origin (clamping or manipulation). Using a rapid immunochemiluminometric assay (ICMA), data were collected from 22 patients successfully operated for parathyroid adenoma. The check at clamping correctly diagnosed 13 patients. Among the 9 false negative cases, 6 were correctly diagnosed considering the manipulation as the baseline value. In the remaining 3 patients, diagnosis required prolonged observation of the curves. In case the iPTH decay does not follow the expected curve, it can be useful to check the decay normalising to a pre-excision value. The advantages of our criterion are both the prompt recognition of false negative results and the construction of a “true” decay curve for each patient, supporting the surgeon during the excision of hyperfunctioning parathyroid tissue.

Key words: Intraoperative PTH, Primary hyperparathyroidism, PTH decay, Time origin shift

IN ORDER to improve surgical cure of primary hyperparathyroidism, a rapid assay for intact parathyroid hormone (iPTH) to be used intraoperatively was developed [1].

The PTH monitoring is based on the assumption that, when the affected gland is excised, the steady state is altered and PTH begins to decrease. Thus, the instant of the excision is considered the origin of the decay curve and the cut-off is defined as a percentage of the origin iPTH level measured few minutes afterwards.

Nussbaum et al. demonstrated a decline of PTH to less than 40% of the baseline values (just before legation of the vascular pedicle) 15 min after successful parathyroid adenomectomy in 12 patients [1].

Subsequently, other Authors checked the cut-off earlier (10 min after the excision) and, obviously, since the curve is decreasing, they considered a higher cut-off value (50%). These criteria have good results when the instant of the excision is truly the time origin of the decay [2]. However, iPTH measurements between the pre-incision and pre-excision moments were shown to increase as a consequence of unavoidable surgical manipulations [3]. Furthermore, the PTH measurement at the excision time could be very low if the surgical manoeuvres for isolation of the parathyroid adenoma compromise the vascularization and provoke ischemia of the adenoma. Therefore, both false negative and false positive results have occurred in the interpretation of the intraoperative iPTH test. These pitfalls prompted the modification of the originally proposed methodology of evaluation [2, 4-9]. Table 1 reports the seven main criteria that have been proposed taking into account also PTH values during the pre-incision/ pre-excision interval [10, 11].

In the last years the most applied of these methods...
patients (C1-C20) successfully operated for single parathyroid adenoma [Ref. 12 Fig. 2; http://www.eje-online.org/cgi/reprint/144/4/353]. All but two patients had a decay curve made by two components: the first with a rapid decrease lasting about 5 minutes and the second with a slow decrease lasting about 15 minutes. In these patients the iPTH values taken at 10 minutes after clamping are under 50% of the basal level. The iPTH at 10 minutes after clamping was not decreased in 2 patients who were cured by the excision of a single adenoma (false negative results). Therefore, in order to understand the reason of these two false negative results, we supposed that sometimes the origin of the curve might not coincide with the instant of the announced clamping, but with a preceding instant. A mathematical simulation of this hypothesis was then tested. Taking into account the decay curve (from clamping) of patient C4 (our best fitting among all the 20 Pts; r²≈1), other curves were simulated using the same parameters, but with earlier origins. In Fig.1 the curve labelled “0 min” is just this curve and its origin coincides with the origin of the axes. The three simulated curves originate respectively 3, 6, 9 minutes before the 0 minute and are drawn as dotted in their first part (before time 0) and as solid after time 0. If the solid parts of these curves are normalized at their maximum value, that is at time 0, the trend is completely different as it is shown in the inset of Fig. 1. Quite surprisingly the trend of the curves designed at -6 and -9 minutes is similar to that of two patients (C17 and C19) having a false negative response of iPTH assay. On the contrary, the -3 minutes curve results less modified and it could indicate a curative surgery.

Fig. 2 shows how the curves of patients C17, C19 can be actually simulated by shifting the curve of C4, 10 min earlier for C17 and 7 min for C19. Since these time frames are consistent with the usual exposure time of the adenoma, the right origin of iPTH decay

has been the Miami criterion that considers as basal level the highest PTH value obtained before the adenoma excision. Surprisingly the cut-off value was always checked at 10 min after the excision, not considering the time elapsed from this new baseline that could be extremely variable [4, 7-9].

The timing of the sample that can result as “the highest value” is not well defined and its relationship with the other samples is never specified. However, the information given by an intermediate value seems too important to be neglected and, in our study, an attempt to properly use it was made by supposing the coincidence of value taken during the manipulation phase with the origin of the iPTH decay.

Material and Methods

a) Theoretical considerations

The iPTH kinetics during surgery for parathyroid adenoma is well described by a two-compartment (circulating blood and extravascular) model [12]. The serum iPTH decay after the removal of the parathyroid adenoma is represented by a curve expressed by two exponential functions whose sum results in two phases: its first phase (fast) is due to the sudden lack of iPTH after parathyroid adenoma resection; the second phase is influenced also by the incretion of the other parathyroid glands (either normal and unsuppressed or pathological) and therefore it shows a slower decrease. The mathematical expression for calculating the circulating iPTH decay curve at every time (t), after the parathyroid adenoma excision, is the following:

$$[\text{iPTH}(t)] = A e^{-at} + B e^{-bt} + EV$$  \hspace{1cm} (Eq.1)

where t is the time from the decay origin, $A e^{-at}$ is the fast phase, $B e^{-bt}$ is the slow phase of decay (being $a > b$) and $EV$ (equilibrium value) depends on the contribution to PTH incretion from the other parathyroid glands. We had verified our theoretical model in twenty

| Miami Criterion: >50% drop from the highest iPTH level 10 min after gland excision. |
| Criterion 1 or Vienna Criterion: >50% drop from the pre-incision iPTH level 10 min after gland excision. |
| Criterion 2: >50% drop from the highest iPTH level 10 min after gland excision and a final iPTH level within the reference range. |
| Criterion 3: >50% drop from the highest iPTH level 10 min after gland excision and a final iPTH level lower than the pre-incision value. |
| Criterion 4: >50% drop from the highest iPTH level 5 min after gland excision. |
| Criterion 5: >50% drop from the pre-excision iPTH level 10 min after gland excision. |
| Halle Criterion: iPTH decay into the low normal range (≤ 35 ng/L) within 15 minutes after gland excision. |

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True time origin of iPTH decay curve

b) Clinical data
In this paper, new twenty two patients (P1-P22) with primary hyperparathyroidism were successfully operated for single gland disease by minimally invasive adenectomy performed by the same surgeon during the year 2011. They were 15 females and 7 males.

is likely to coincide in these cases with the instant of manipulation, i.e., when the surgeon greatly affects the gland. In practice, such a situation may happen when the squeezing of the gland has caused a rapid efflux of the hormone. Having accepted our hypothesis and the criterion of comparison with the iPTH level at origin, the measurement of the time interval (10 min) for the second sample must be computed starting from this new origin and no longer from the excision instant.

b) Clinical data
In this paper, new twenty two patients (P1-P22) with primary hyperparathyroidism were successfully operated for single gland disease by minimally invasive adenectomy performed by the same surgeon during the year 2011. They were 15 females and 7 males.
with a mean age 51.8 years (range 43-73). All patients remained normocalcemic 6 months after parathyroidectomy (normal value 8.2-10.7 mg/dL). Their records were characterized by peripheral blood collected at anaesthesia induction, during the isolation of the suspected hyperfunctioning gland that the surgeon judged as the phase of maximal manipulation (this time was usually few minutes - mean 8’ 20"- before the parathyroid adenoma excision), at clamping, 10 min after the sample indicated as manipulation, 10 min after clamping, and onwards until required (usually 20 and 30 minutes after clamping) (Table 2). Data were obtained using the iPTH immunochemiluminometric assay (ICMA) (Quick-Intraoperative™ intact PTH, Nichols Laboratories, San Juan Capistrano, California, USA), performed outside the operating room using a portable analyser. To our knowledge, this assay is, at the present, unavailable but this is unessential because all the curves are normalized and, thus, any good assay can be used.

Table 2 iPTH data (pg/mL) in 22 patients successfully operated on for single adenoma

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*iPTH*<sub>BV</sub> = iPTH at anesthesia; *iPTH*<sub>man</sub> = at manipulation; *iPTH*<sub>cl</sub> = iPTH at clamping.
In parentheses, minutes after clamping (before) and minutes after manipulation (behind).
c) Statistical and graphical analysis

Statistical and graphical analysis were carried out using the package Statistica for Windows (Statsoft Inc., Tulsa, Oklahoma, USA) and MATLAB macrolanguage (Math Works Inc., Massachusetts, USA), implemented on an IBM-compatible personal computer (Windows XP).

Results

All the 22 curves, linearly interpolated and normalized to the clamping value (the standard origin), are reported in Fig. 3. In single adenoma, the patients were expected to have a >50% drop from the clamping value at 10 min after it. However, only 13 patients had this result, whereas 9 patients showed a <50% drop. Therefore, the latter patients could be categorized as not cured.

According to our hypothesis, if the check versus clamping value shows a decrease < 50%, the time origin may have coincided with the gland manipulation and, therefore, a new check must be performed taking into account this alternative origin. Obviously, in this case, the value at clamping (no longer a “total”, but clearly a “partial” clamping) must be an intermediate point of the curve and lower than the value at manipulation. Since we stated the check at 10 min, only those patients with the “manipulation-clamping” time interval (Tmc) ≤ 10 min could be considered for the second check. Of the 9 patients with false negative result (P2, P3, P5, P10, P11, P13, P14, P17, P21), 6 fell under this condition, whereas 3 did not (P10, P11, P14). The 6 curves, linearly interpolated and now normalized to the manipulation value, showed a ≥ 50% drop from it (Fig. 4). For the 3 patients with Tmc > 10 min, the extended observation of the decay curves by additional sampling correctly diagnosed them as cured.

In Fig. 5 are reported the curves of the 19 correctly diagnosed patients evaluated either versus clamping (n.13) or versus manipulation (n.6) with analogous decay. Moreover, we drew a curve linearly-interpolated representing the upper limit of above mentioned curves, i.e. a cut-off curve. It is noteworthy that all the curves, in accordance to the results obtained by Nussbaum et al. [1], fall under 40% of the origin level at 15 min after it, which means that the searching for time origin allows to draw (though approximately) the kinetics of each patient and shows how the cut-off must be modified with regard to the time interval cho-

![Figure 3](https://via.placeholder.com/150)

**Fig. 3** Percentage changes of iPTH, linearly interpolated from clamping in 22 patients with single adenomas. Solid lines: curves of 13 patients correctly diagnosed (true positive results); dashed lines: curves of 9 patients erroneously diagnosed (false negative results).
Fig. 4  Percentage changes of iPTH, linearly interpolated from manipulation in 6 patients (with Tmc ≤ 10 min) resulted false negatives at the first check. The full circles mark the clamping values.

Fig. 5  Percentage changes of iPTH, linearly interpolated from the “most probable origin” of the 19 patients correctly diagnosed by the two checks, either versus clamping (13 patients, thin solid lines) or versus manipulation (6 patients, dashed lines). The curves fall under the alternative cut-off of Nussbaum too (40% at 15 min). Thick line represents the linearly interpolated limit curve (representing the values under which all the 19 curve values are contained).
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Discussion

Protocols which provide the comparison between the PTH level at clamping and 10 minutes later refer to at least one of the following hypothesis: 1) the excision time (clamping) is rigorously the starting point of the decay curve; 2) the decay curve is described by a simple exponential function.

All the above mentioned iPTH monitoring criteria (Table 1) are based on the definition of a cut-off value and on the mere comparison between only two values; both the cut-off value as well as the time interval between the pre- and the post-excision points are empirically defined. Our study, on the contrary, addresses one of the main problems presented in the literature, i.e. the choice of the value to which the post-excision sample has to be compared, and it proposes a slightly modified method in order to increase the probability of finding the temporal origin of the curve.

If the decay curves were described by a mono-exponential function, the problem would not exist (Fig 6a). Such a situation, according to our experience, can occur rarely, since it only simulates the presence of a single adenoma with the other glands totally suppressed in the absence of any parenchymal exchange. In this case, it is well known that a “half-time” can be defined regardless of the position of the two points on the curve. Consequently, establishing the 50% cut-off at 10 min, it would be sufficient to measure two samples in a time frame equal to 10 min and to check whether the second value is ≤50% of the first. Nevertheless, our previous study [12] and the one by Maier and coll. as well [13] showed that more reliable results can be obtained describing the curve by a bi-exponential function plus a base level (Eq. 1). Consequently, the term “half-time” becomes meaningless since the function is halved at longer successive intervals. It must, therefore, be taken into account only the first halving, calculated starting from the time origin of the decay curve (truly “basal point”) (Fig. 6b).

As many different situations may occur during the exposition of the gland, it is our opinion that the instant of manipulation cannot be established a priori, but the surgeon should identify, during the exposure of the adenoma, the moment when the hormonal release is likely to be greatly influenced. Obviously, the manipulation may not always be the right origin when the check at clamping fails, but this is not relevant, since “manipulation check” does not replace the “clamping check”, being included in order to increase the probability of finding the true origin.

Most of the other criteria select the check at manipulation when it is the highest value (Miami Criterion and Criteria 2, 3, 4), but not assuming it as the origin. Consequently, iPTH at 10 min from it is not measured, using for comparison always the value at 10 min from the excision. Even if sometimes the results can be equally valid, the theoretical basis for this interpre-

Fig. 6 Sect. a) Decay curve of a mono-exponential function with a half-time of approximately 7 minutes. It is evident that there is no need to know what is the starting point, because the value halves always after the same interval. Sect. b) Decay curve of our model: the initial half-time (~10 min) increases over time (~18 min); as a consequence, the first half-time can be rightly calculated only if the curve origin is known.
tation is lacking.

At first observation our method could seem laborious but its application requires only one more sample for a simple calculation, i.e. the check at 10 min after manipulation. Consequently, the added “cost” is negligible when compared to the “benefit”. In this study we immediately resolved the problem of 6 out of the 9 patients diagnosed as not cured at the check from clamping, with the imaginable pros. However, to obtain an alternative origin with the highest probability, a greater care is required both in the timing of sampling and in the recognition of the critical manipulation time. In some cases the origin cannot be clearly recognized, or the above mentioned Tmc is longer than 10 min, as it happened in 3 out of the 22 patients considered in this study, and the observation of the curve must be elongated, by examining subsequent blood samples taken 20 and 30 minutes after the parathyroid adenoma excision.

The main advantage of the recognition of the time origin (clamping or manipulation) is not the “direct recovery” of false negative results, but the possibility of constructing the approximate decay curve for each patient (i.e. the personalized kinetics), a task not easily derived by other methods [3]. Interestingly, being the curves derived from a homogeneous population, statistical analyses can be performed, with percentile interpretation that acquires higher significance when increasing the number of observed cases in the chosen cohort.

Another important goal in iPTH monitoring is the minimization of false positive results. At present, false positive results represent variable percentages of total analyses, depending on the adopted criterion [11]. However, with a normal population of data, a “cut-off curve” equivalent, for instance, to 90° or 85° percentile could be analytically derived, with the possibility of setting the cut-off value in a statistically controlled manner. In other words, the probability level could be established a priori and consequently the cut-off curve could be calculated. A lower cut-off value would obviously lessen the risk of false positive results though increasing the risk of false negatives that, however, may be easily controlled by a prolonged observation of the curve.

Recently, Riss et al. analyzed, in patients operated on for primary hyperparathyroidism, the risk factors for the presence of spikes during intraoperative iPTH monitoring using different criteria (Miami, Vienna, Halle), observing for all these criteria an increased risk of false negative results. The Authors conclude calling for the identification of a new method to optimize the iPTH monitoring, through an individual interpretation of the iPTH decay curve [14]. In our opinion, the performance of a blood sample at the time of adenoma manipulation and the iPTH check after an interval of about 10 min helps to reduce the incidence of false negative results, avoiding the unnecessary prolongation of cervical exploration.

In conclusion, the results of our analysis clearly point to the need of finding the “true”, or at least “the most probable”, time origin in order to improve the quality of the intraoperative PTH monitoring.

Acknowledgments

This work was supported by the ordinary funding from the Italian Government.

Disclosure statement

The authors have nothing to disclose. No Conflicts of Interest were present and Ethical Adherence was applied. The authors declare that the manuscript is not submitted to any other journal.

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