Pediatr Transplantation 2015: 19: E37–E40

Pediatric Transplantation
DOI: 10.1111/petr.12410

A novel non-surgical, minimally invasive technique for parathyroid autotransplantation: A case report

Aysan E, Kilic U, Gok O, Altug B, Ercan C, Idiz UO, Kesgin C, Muslumanoglu M. (2015) A novel non-surgical, minimally invasive technique for parathyroid autotransplantation: A case report. *Pediatr Transplant*, 19: E37–E40. DOI: 10.1111/petr.12410.

Abstract: We present a case report of intramuscular autotransplantation of the parathyroid cell suspension acquired after total parathyroidectomy. A 15-yr-old female patient who had been undergoing hemodialysis due to chronic renal failure for eight yr was diagnosed with secondary hyperthyroidism and subsequently underwent total parathyroidectomy. The parathyroid cells were acquired from the resected tissues, processed through isolation and cultivation phases, and counted using a cell counter. A total of two million cells were injected into the left deltoid muscle using a 22-gauge needle. After surgery, five and 10 million cells were injected in the fifth and 12 week, respectively. The desired serum levels of parathyroid hormones and calcium were not achieved after the first two transplantations. In addition, there was no regression in the patient's symptoms. However, at four wk after the third transplantation, serum parathyroid hormone level did not decrease to <3 pg/mL, the patient was asymptomatic, and the oral treatment was stopped. Our findings indicate that this new technique is applicable because it is minimally invasive, and it can be easily repeated.

Erhan Aysan[1], Ulkan Kilic[2], Ozlem Gok[2], Burcugul Altug[2], Cilem Ercan[2], Ufuk Oguz Idiz[1], Cemile Kesgin[1] and Mahmut Muslumanoglu[1]

[1]Department of General Surgery, Faculty of Medicine, Bezmialem Vakif University, Istanbul, Turkey, [2]Medical School, Bezmialem Vakif University, Istanbul, Turkey

Key words: autotransplantation – invasive – minimally – non-surgical – parathyroid

Ufuk Oguz Idiz, Ihlas Marmara Evleri 1, Kisim A-18 Blok Daire: 5, Beylikduzu, Istanbul 34900, Turkey
Tel.: +905062044714
Fax: +902122240772
E-mail: oguzidiz@yahoo.com

Accepted for publication 13 November 2014

The treatment of renal hyperparathyroidism is continuously evolving. Patients with hyperparathyroidism secondary to CKD are sometimes treated with subtotal parathyroidectomy (1, 2). Several non-invasive treatment methods that reduce the PTH, increase the calcium, and decrease the bone turnover have been developed for patients with CKD. However, some cases may require surgical intervention (3–6).

There are two fundamental approaches in the surgical treatment of secondary hyperparathyroidism in patients with CKD: subtotal parathyroidectomy or total parathyroidectomy with autotransplantation. The second technique is preferred because it is easy and carries a low risk of complication in case of recurrence. Autotransplantation, coined by Wells, involves implanting the resected parathyroid tissue into the muscle – usually the forearm muscles (7).

In this study, we present the results of our new minimally invasive parathyroid transplantation technique performed on a 15-yr-old patient who had total parathyroidectomy due to secondary hyperparathyroidism.

Methods

The 15-yr-old female patient with CKD who had been receiving hemodialysis for eight yr was referred to the Bezmialem Vakif University Department of General Surgery, Field of Endocrine Surgery outpatient clinic with the diagnoses of treatment-resistant secondary hyperparathyroidism. The patient had been using sevelamer hydrochloride (Renagel® Sanofi-Aventis, Istanbul, Turkey) 2 × 800 mg for more than two yr, calcium carbonate (Antifosfat CC®; Assos Pharmaceuticals, Istanbul, Turkey) 6 × 500 mg, and cinacalcet HCl (Mimpara®, Istanbul, Turkey) 1 × 60 mg. Serum PTH > 1900 pg/mL, and calcium was 9.3 mg/dL.

Abbreviations: CKD, chronic kidney disease; FBS, fetal bovine serum; PBS, phosphate-buffered saline; PTH, Parathyroid hormone.

The patient received an operation preparation with the purpose of autotransplantation and a total parathyroidectomy. The Bezmialem Vakif University local human ethic committee approved this study. Because the patient was under 18 yr of age, both she and her mother were orally informed of the risks and benefits of the study and their written consents were obtained.

Total parathyroidectomy was performed using a standard Kocher incision with the patient under general anesthesia. One half of all the excised parathyroid tissues were sent to the pathology department for histopathologic evaluation, and the remaining half were sent to parathyroid transplantation laboratory kept in a culture medium, which is composed of 4 mL of AmnioMAX™ II Complete Medium (cat. no: 11269-016; Gibco Life Technologies, Carlsbad, California, USA) with 20% inactivated FBS (cat. no: 10500-064; Gibco Life Technologies) and 1% penicillin–streptomycin (cat. no: 15140-122; Gibco Life Technologies) in a melting-ice solution at 4 °C for subsequent culturing immediately. After confirmation response of hyperplasia of the tissues from pathological examination, our new cell culture and preparation protocol was implemented. The main parts of the resected tissues were washed with the culture medium five times. The collected tissues were isolated from blood vessels, gland capsule, connective and fatty tissues. Remaining parathyroid tissue pieces were mechanically disintegrated using a sterile filter in 1× PBS (cat no: AM9624; Ambion Life Technologies, Carlsbad, California, USA) supplemented with 5% inactivated FBS under sterile conditions in a biohazard safety cabinet (cat. no: L.02131262, Mars Safety Class 2, SCANLAF, Lynge, Denmark). Subsequently, the solution was filtered into a 15-mL tube using a sterile cell strainer (100 μm, cat.no: 352360, Falcon, BD Biosciences, NJ, USA) and mixed with 600 μL deoxyribonuclease I (from bovine pancreas, A3778-0010; AppliChem, Darmstadt, Germany). After centrifugation at 200 × *g*. for five min at room temperature, the supernatant was removed and the pellet was rapidly suspended in 1 mL of culture medium. The cell viability was 85.79% as assessed by Muse™ Cell Analyzer (cat.no: 0500-3115 Merck Millipore Darmstadt, Germany). The counted cells (approximately 50% parathyroid cells of 320×10^6 cells) were used for cultivation. The cell suspension was cultivated in two flasks (cat.no: sc-200262,UltraCruz™, Santa Cruz Biotechnology, Dallas, TX, USA), each containing 10 mL of the culture media and kept in the incubator (CCL-170B-8; ESCO, Singapore City, Singapore) at 37 °C, 5% CO_2-containing humidified atmosphere for 24 h. The upper medium in one of the flasks was transferred into a 15-mL tube, the cellular monolayer was washed with 4 mL PBS, and then, 300 μL trypsin (25300-054; Gibco, UK) was added to the flask and kept in the incubator to detach the cells from the flask for five min and transferred into a tube. After centrifugation at 200 × *g* for five min, the supernatant was removed and the pellet was suspended in 1 mL of cell culture media; 145×10^6 living cells in 1 mL culture media were detected.

The cell suspension was transferred into a microcentrifuge tube and centrifuged at 200 × *g* for five min. The pellet was suspended in 2 mL of the recipient's blood serum for cell transplantation. The rest of cells were then cryogenically stored in culture media after mixing with 5% DMSO (cat.no: sc-202581, ChemCruz™, Santa Cruz Biotechnology) using a controlled-rate freezer.

To avoid recurrence of hyperparathyroidism, the process of autotransplantation was performed gradually, increasing number of cells. At the second postoperative week, we injected two million cells in the left deltoid muscle using 22-gauge needles into a total volume of 2 mL. In the follow-ups after the first transplantation, the patient's symptoms were not lost, and no increase was detected in her PTH and calcium values. Thus, the patient was injected five million cells in the seventh week. Upon non-detection of clinical and biochemical recovery in the follow-ups of the patient, the patient was injected 10 million cells postoperatively in the 12th week. We observed the patient in the outpatient clinic.

Results

There were no complications following the total parathyroidectomy. The patient was discharged on postoperative day two with oral calcium (Calcium Sandoz efervesan tablet®; Eczacibasi Co., Istanbul, Turkey) 3 g/day. The change in the levels of serum PTH and calcium from before the operation to after the third autotransplantation is shown in Figs. 1 and 2. Serum PTH did not drop below 3 pg/mL, and serum calcium increased above 7 mg/dL four wk after the third autotransplantation. In this period, calcium levels did not decrease, and the patient's symptoms such as tingling, muscle aches and cramps, fatigue, and weakness disappeared. Thus, additional treatments were deemed unnecessary; however, because of the hungry bone syndrome, we continued regular outpatient follow-up.

Conclusion

The incidence of symptomatic hyperparathyroidism is high in patients with CKD (8). According to the European Dialysis and Transplant Association, the incidence of parathyroidectomy in renal patients is about five per 1000 patients per year during the first 2–3 yr of dialysis (9). A total of 15% and 38% of patients with CKD receiving dialysis for 10 and 20 yr, respectively, require parathyroidectomy (10). Parathyroidectomy is usually performed in patients who cannot tolerate or are resistant to medical treatment and who lack sufficient regression in levels of PTH, calcium, and phosphorus despite the use of calcitriol, cinacalcet, and other vitamin D analogs (11).

The first study on parathyroid autotransplantation was performed by Halsted in 1907, and he achieved successful results in 61% of the dogs on which he performed autotransplantation to the rectus muscle and thyroid gland (12). Lahey was the first person to apply parathyroid autotransplantation to humans (13).

Different anatomic regions of the body are preferred for the parathyroid tissue autotrans-

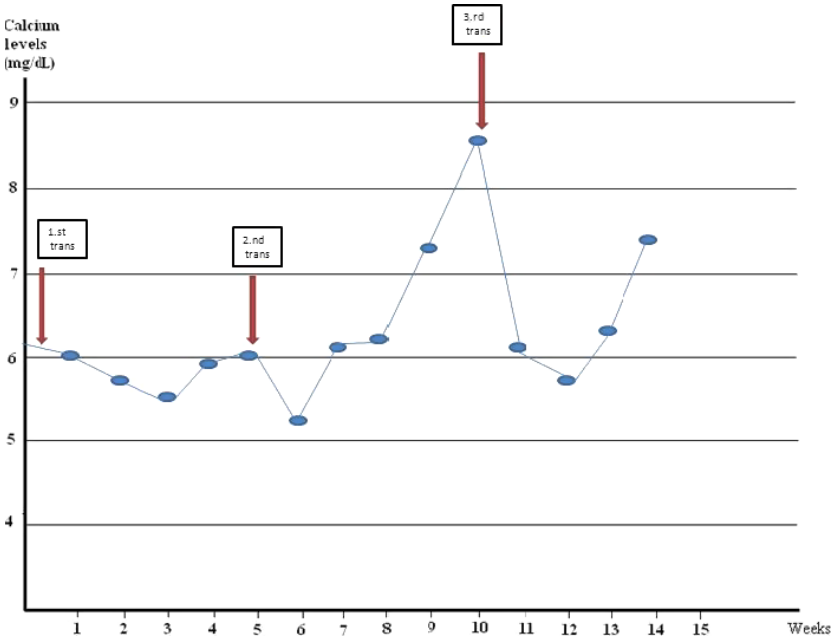

Fig. 1. The change in the values of serum calcium from before the operation to after the third autotransplantation.

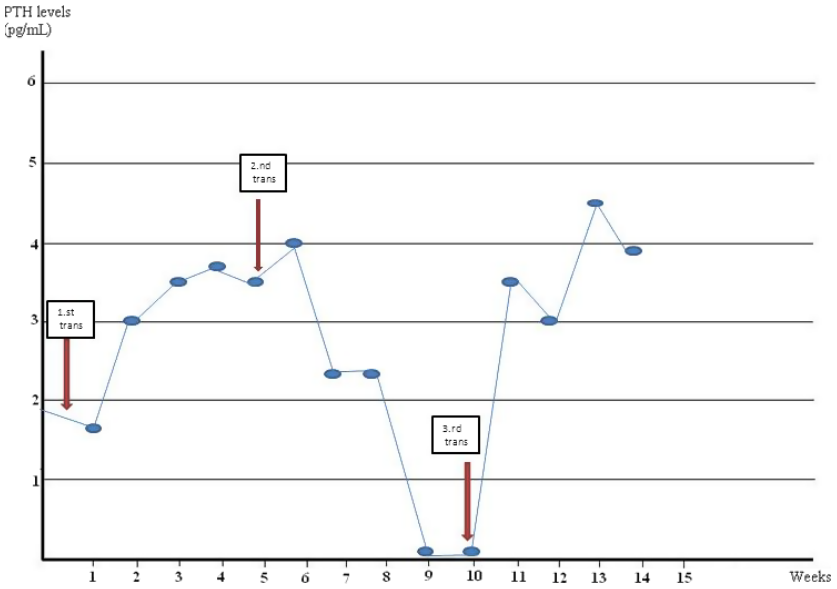

Fig. 2. The change in the values of serum PTH from before the operation to after the third autotransplantation.

plantation, such as the abdominal region, parasternal region, subcutaneous tissue of the forearm, and sternocleidomastoid muscle (8, 14–17). However, the most commonly preferred method is the parathyroid tissue autotransplantation performed between the forearm muscles, first described by Wells et al. (7). This method is widely used because the forearm is easily accessible. However, both Wells' and the other autotransplantation techniques are invasive and lead to scar development and complications such as anatomic restriction (18).

In our study, parathyroid autotransplantation was carried out by intramuscular injection of a cell suspension after processing cells through isolation and cultivation phases and counting them with a cell counter. To the best of our knowledge, this is the first report of such a method. A similar approach was performed by Tan et al.; however, in that study, the tissue was injected intramuscularly with a wide injector after being segmented, so the phases of cell isolation, cultivation, cell counting, and cell suspension were not used (19). In the present study, a minced tissue injection technique was compared with the standard surgical technique, the result was 69% with the injection group and 87% with the standard surgical group, and the difference was not

significant. The recurrence rate was 12.9% in the standard surgical group and 2.9% in the injection group.

In previous studies, fetal or adult bovine serum and antibiotics similar to our culture environment were used and, as in our study, no allergic reactions and complications were encountered (20, 21).

The advantages of our technique are as follows: It does not require an incision, it is minimally invasive and easily applicable, and cell injections can easily be repeated in case of treatment failure. In fact, we performed the first autotransplantation using two million cells, which failed, following which we increased the number of cells to five million and eventually to 10 million.

Because the clinical and biochemical results that were expected from the first two transplantations were not obtained, we needed to increase the number of cells in the third transplantation. The success of the final transplantation could explained by one or more of the following reasons: the number of cells administered, enhanced cell-to-cell contacts that extend the lifetime of the cells, the two previous transplantations being unsuccessful due to the hungry bone syndrome occurring prior to transplantation, and due to saturation of the bones following the first two transplantations.

A disadvantage of our technique is that it requires a cell culture laboratory with the technical expertise to perform the cryopreservation process. Furthermore, the long-term results of this new technique are not clear, and further studies are required to clarify this. Nevertheless, our findings show that short-term results are satisfactory, and it is possible to repeat autotransplantation with the desired number of parathyroid cells.

Disclosure

Erhan Aysan and all the other authors agreed on the content of the article as it is. I also declare that there is no conflict of interest and no form of support such as grant, equipment, and/or pharmaceutical items for the article.

References

1. Al-Azem H, Khan AA. Hypoparathyroidism. Best Pract Res Clin Endocrinol Metab 2012: 26: 517–522.
2. Glockzin G, Hornung M, Kienle K, et al. Completion thyroidectomy: Effect of timing on clinical complications and oncologic outcome in patients with differentiated thyroid cancer. World J Surg 2012: 36: 1168–1173.
3. Llach F. Secondary hyperparathyroidism in renal failure: The trade-off hypothesis revisited. Am J Kidney Dis 1995: 25: 663–679.
4. Akizawa T, Fukagawa M, Koshikawa S, et al. Recent progress in management of secondary hyperparathyroidism of chronic renal failure. Curr Opin Nephrol Hypertens 1993: 2: 558–565.
5. Fournier A, Moriniere PH, Oprisiou R, et al. 1-Alpha-hydroxyvitamin D3 derivatives in the treatment of renal bone diseases: Justification and optimal modalities of administration. Nephrology 1995: 71: 254–283.
6. Llach F. Parathyroidectomy in chronic renal failure: Indications, surgical approach and use of calcitriol. Kidney Int Suppl 1990: 29: 62–68.
7. Wells SA Jr, Gunnels JC, Shelburne JD, et al. Transplantation of parathyroid glands in man: Clinical indications and results. Surgery 1975: 78: 34–44.
8. Monchik JM, Bendinelli C, Passero MA Jr, Roggin KK. Subcutaneous forearm transplantation of autologous parathyroid tissue in patients with renal hyperparathyroidism. Surgery 1999: 126: 1152–1159.
9. Wing AJ, Boyer M, Brunner FP, Brynger H, Tufverson G, Selwood NH. Combined report on regular dialysis and transplantation in Europe. XV 1984. Proc Eur Dial Transplant Assoc 1985: 22: 5–54.
10. Fassbinder W, Brunner FP, Brynger H, et al. Combined report on regular dialysis and transplantation in Europe. Nephrol Dial Transplant 1991: 6: 5–35.
11. Kidney Disease: Improving Global Outcomes (KDIGO) CKDMBD Work Group. KDIGO clinical practice guideline forthe diagnosis, evaluation, prevention and treatment of chronic kidney disease-mineral and bone disorder (CKD-MBD). Kidney Int 2009: 113(Suppl): S1–S130.
12. Halsted WS. Auto- and isotransplantation, in dogs, of the parathyroid glandules. J Exp Med 1909: 11: 175–199.
13. Lahey FH. The transplantation of parathyroids in partial parathyroidectomy. Surg Gynecol Obstet 1926: 62: 508.
14. Chou FF, Chan HM, Huang TJ, et al. Autotransplantation of parathyroid glands into subcutaneous forearm tissue for renal hyperparathyroidism. Surgery 1998: 124: 1–5.
15. Kinnaert P, Salmon I, Decoster-Gervy C, et al. Long-term results of subcutaneous parathyroid grafts in uremic patients. Arch Surg 2000: 135: 186–190.
16. Jansson S, Tisell LE. Autotransplantation of diseased parathyroid glands into subcutaneous abdominal adipose tissue. Surgery 1987: 101: 549–556.
17. Lieu D, Hirschowitz SL, Skinner KA, Zuckerbraun L. Recurrent secondary hyperparathyroidism after autotransplantation into the sternocleidomastoid muscle: Report of a case with fine needle aspiration findings. Acta Cytol 1998: 42: 1195–1198.
18. Yoon JH, Nam KH, Chang HS, et al. Total parathyroidectomy and autotransplantation by the subcutaneous injection technique in secondary hyperparathyroidism. Surg Today 2006: 36: 304–307.
19. Tan CC, Cheah WK, Tan CT, Rauff A. Intramuscular injection of parathyroid autografts is a viable option after total parathyroidectomy. World J Surg 2010: 34: 1332–1336.
20. de Menezes Montenegro FL, Custódio MR, Arap SS, et al. Successful implant of long-term cryopreserved parathyroid glands after total parathyroidectomy. Head Neck 2007: 29: 296–300.
21. Cabané P, Gac P, Amat J, et al. Allotransplant of microencapsulated parathyroid tissue in severe postsurgical hypoparathyroidism: A case report. Transplant Proc 2009: 41: 3879–3883.