Int J Clin Exp Med 2015;8(3):4611-4619
www.ijcem.com /ISSN:1940-5901/IJCEM0004971

Case Report

Replantation with cryopreserved parathyroid for permanent hypoparathyroidism: a case report and review of literatures

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Received December 17, 2014; Accepted February 11, 2015; Epub March 15, 2015; Published March 30, 2015

Abstract: Permanent postsurgical hypoparathyroidism is defined as insufficient parathyroid hormone (PTH) to maintain normocalcemia 6 months after surgery. It occurs mostly in reoperation for persistent or recurrent hyperparathyroidism. The treatment of long-term calcium and vitamin D supplement is burdensome and may cause iatrogenic complications. PTH replacement is potential but still under trials. Only replantation with cryopreserved parathyroid is an available treatment for patients to reduce or stop long-term drug administration. However, this treatment is not applied widely in developing countries, due to lack of experiences and skills. Herein, we reported a 58-year-old male presented a continuous elevated parathyroid hormone up to about 2342 ng/L and bone pain during hemodialysis for 6 years due to chronic renal failure. He underwent the first operation total parathyroidectomy and autotransplantation. After this operation, he suffered from a persistent calcemia and permanent hypoparathyroidism. After three times of replantation with cryopreserved parathyroid and dialysis with a high calcium dialysate, the low concentration of calcium was elevated and symptoms of hypocalcemia disappeared. However, PTH was not elevated significantly in the long term. It might be related to our nonstandard cryopreservation protocol and no microbiological and histological examinations before replantation, compared with other successful reports. Therefore, we suggest a standard cryopreservation protocol should be followed by non-experienced institutions, especially in developing countries. Furthermore, a high calcium dialysate is efficient to increase calcium concentration and alleviate symptoms of hypocalcemia. It may be an available treatment of persistent hypocalcemia and permanent hypoparathyroidism in dialysis patients.

Keywords: Hypocalcemia, cryopreservation, transplantation, parathyroidectomy, hyperparathyroidism

Introduction

Hypoparathyroidism is the most frequent complication of total parathyroidectomy for renal hyperparathyroidism. Due to long-term bone hungry syndrome, most of them are transient. It can be resolved by medical therapy with oral vitamin D and calcium supplement, intravenous calcium supplement and a high calcium dialysate [1] (**Figure 1**). If it does not recover in 6 months after operation, it is defined as permanent hypoparathyroidism [2]. The current medical treatments of permanent hypoparathyroidism contain calcium and vitamin D supplement. Long-term calcium and vitamin D supplement is a great burden to patients. Parathyroid hormone (PTH) replacement with PTH (1-34) or PTH (1-84) is a potential treatment. A few clinical trials showed that PTH replacement could reduce daily calcium and vitamin D supplement, increase bone mineral density and bone turnover [3-6]. However, it needs to resolve some problems such as long-term effects and side effects by large-scale trials before its clinical application. Its subcutaneous usage also causes inconvenience. Therefore, PTH replacement is not yet available for current treatment of permanent hypoparathyroidism. Replantation with cryopreserved parathyroid is an available choice for permanent hypoparathyroidism. Al-

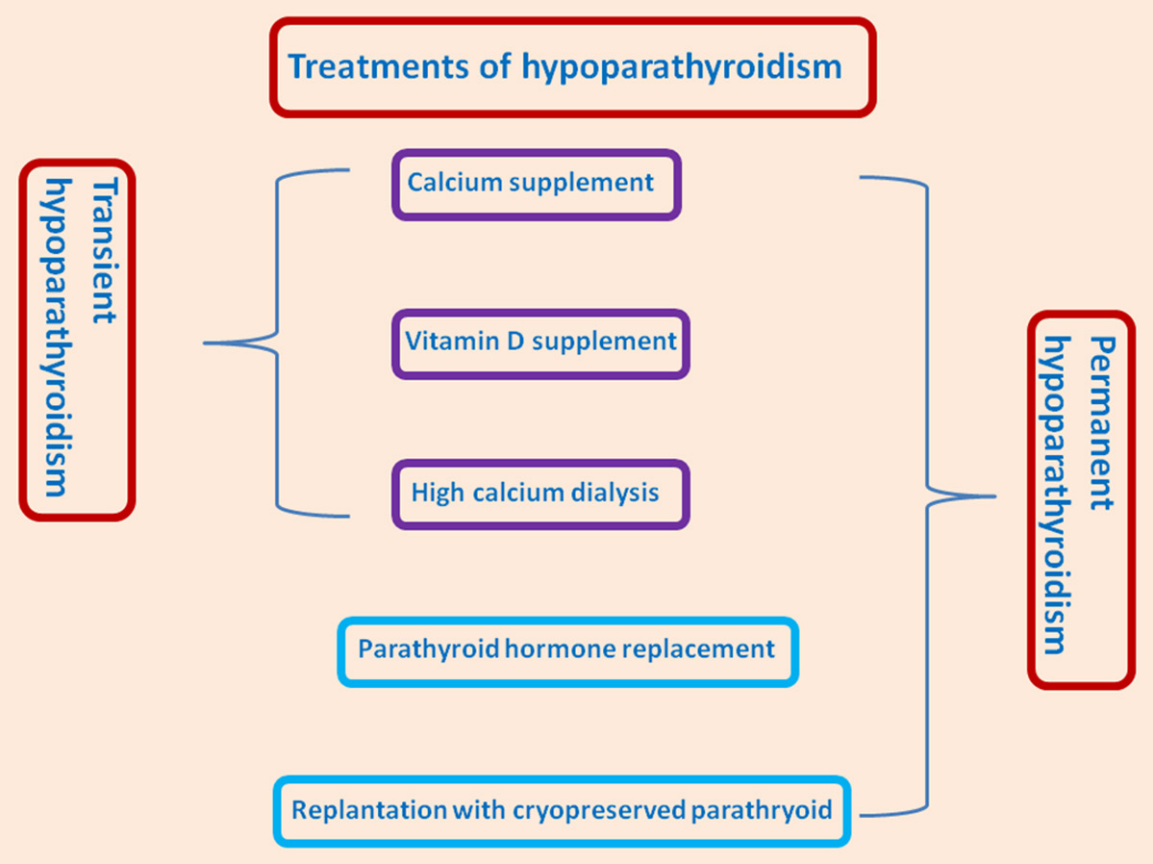

Figure 1. Current treatments of hypoparathyroidism. The treatments of transient hypoparathyroidism contain calcium and vitamin D supplement. High calcium dialysis is only for patients with renal failure. Besides, parathyroid hormone replacement is potential but still needs trials for permanent hypoparathyroidism. Replantation with cryopreserved parathyroid is an available treatment of permanent hypoparathyroidism.

Table 1. Success rates of cryopreserved parathyroid graft

Study	Year	Country	Number	Success rate[&]	The longest duration[*]
Brennan et al [8]	1979	USA	6	67%	18
Wells et al [9]	1980	USA	6	83%	≤ 6
Saxe et al [10]	1982	USA	12	58%	≤ 18
Wagner et al [11]	1991	Germany	25	≥ 64%	84
Herrera et al [12]	1992	USA	12	42%	11
Caccitolo et al [13]	1997	USA	13	46%	≤ 26
Feldman et al [14]	1999	USA	26	58%	125
Cohen et al [15]	2005	USA	26	69%	22
de Menezes Montenegro et al [16]	2007	Brazil	2	-[#]	30
Borot et al [17]	2010	Frence	20	20%	9
Schneider et al [18]	2012	Germany	15	100%	86
Shepet et al [19]	2013	USA	4	25%	< 1
Leite et al [20]	2014	Brazil	1	-[#]	36

[&]: The success rate contain high, fully and partial functional cases. [*]: The longest duration of cryopreservation among high, fully functional or partial functional cases. [#]: Both the studies are case reports.

though first described by Wells et al [7] 40 years ago and followed by many clinics mostly in developed countries (**Table 1**), it is beginning to be conducted in our hospital in last three

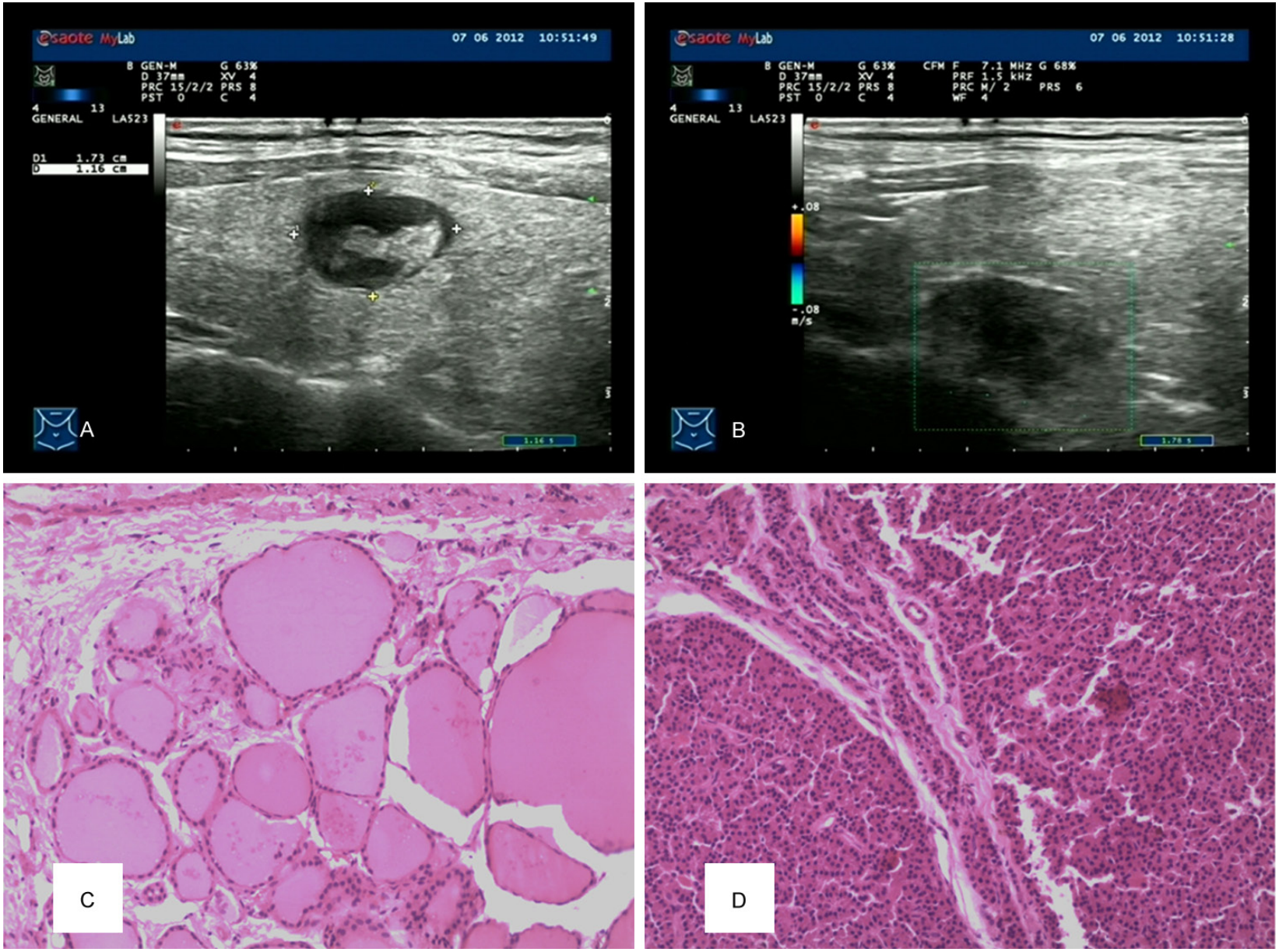
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Figure 2. Images of ultrasound and pathology of the patient. A. A cystic nodule in the right lobe of the thyroid. B. A big parathyroid close to the upper pole of the left lobe. C. The pathological image of nodular goiter. D. The pathological image of parathyroid hyperplasia.

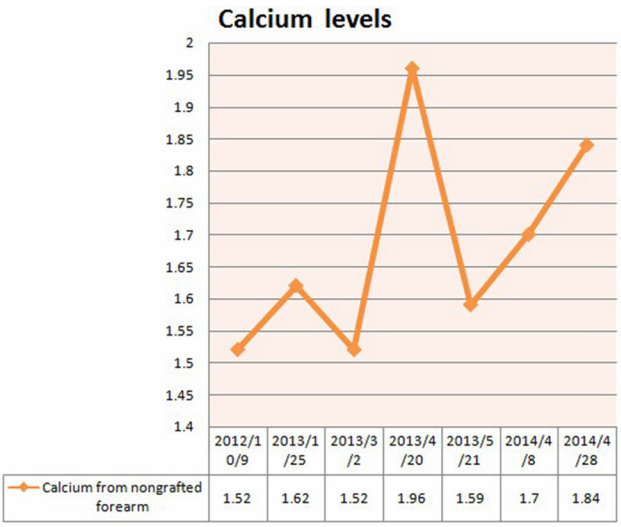

Figure 3. The calcium levels during the treatments.

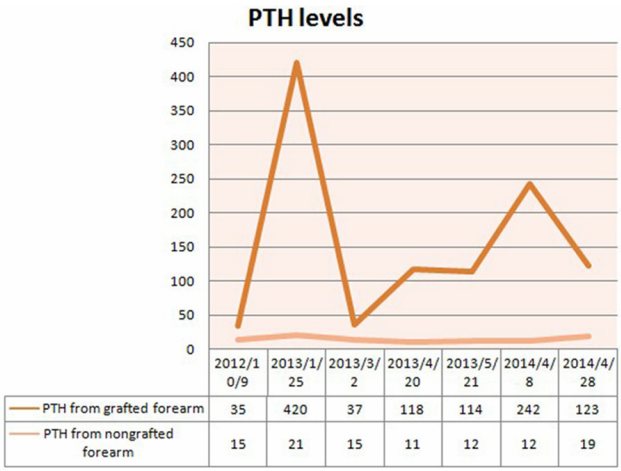

Figure 4. The PTH levels during the treatments.

years. Herein, we reported a case with permanent hypoparathyroidism and hypocalcemia after total parathyroidectomy and autotransplantation for renal hyperparathyroidism. The low concentration of calcium was elevated and symptoms of hypocalcemia disappeared after three times of replantation with cryopreserved parathyroid and dialysis with a high calcium dialysate. However, PTH was not elevated significantly. The causes of failure of our case and factors related to success of cryopreserved parathyroid graft were also discussed.

Case report

A 58-year-old male presented a continuous elevated PTH up to about 1000 ng/L without paresthesias and bone pain during hemodialysis for 6 years due to chronic renal failure. He received a regular hemodialysis three times a week. Meanwhile, he suffered from diabetes more than 8 years and received insulin therapy. He was enrolled in our hospital because he began to have bone and joint pain half a year later with quite a few elevated biochemical markers, PTH 2342 pg/L (normal level 10-69 ng/L), calcium concentration 2.58 mmol/L (normal level 2.10-2.60 mmol/L), phosphorus concentration 2.13 mmol/L (normal level 0.90-1.50 mmol/L), alkaline phosphatase 846 U/L (normal level 53-128 U/L). Ultrasound examination revealed that a hypoechoic and inhomogeneous nodule about 20 × 12 × 12 mm^3 located close to the upper pole of the left lobe, another two similar nodules were close to the inferior pole of the right lobe and three hypoechoic and cystic nodules in the thyroid glands (**Figure 2A** and **2B**).

The first operation: total parathyroidectomy and autotransplantation

He underwent an operation of lobectomy of the right thyroid gland, total parathyroidectomy and transplantation half a parathyroid gland into muscles of the right forearm in July 9, 2012. Meanwhile, the residual parathyroid was cryopreserved. The pathological examination showed thyroid nodular goiters and parathyroid hyperplasia (**Figure 2C** and **2D**). Calcium concentration and PTH deceased immediately after operation. The lowest concentration of calcium was 1.07 mmol/L and PTH was 3 ng/L after several days. Therefore, an aggressive therapy with oral and intravenous calcium supplement was given. He was discharged when calcium concentration increased to 2.1 mmol/L eight days after operation, but PTH was still below normal. He went on to receive oral and intravenous calcium supplement after discharge. Meanwhile, he received a

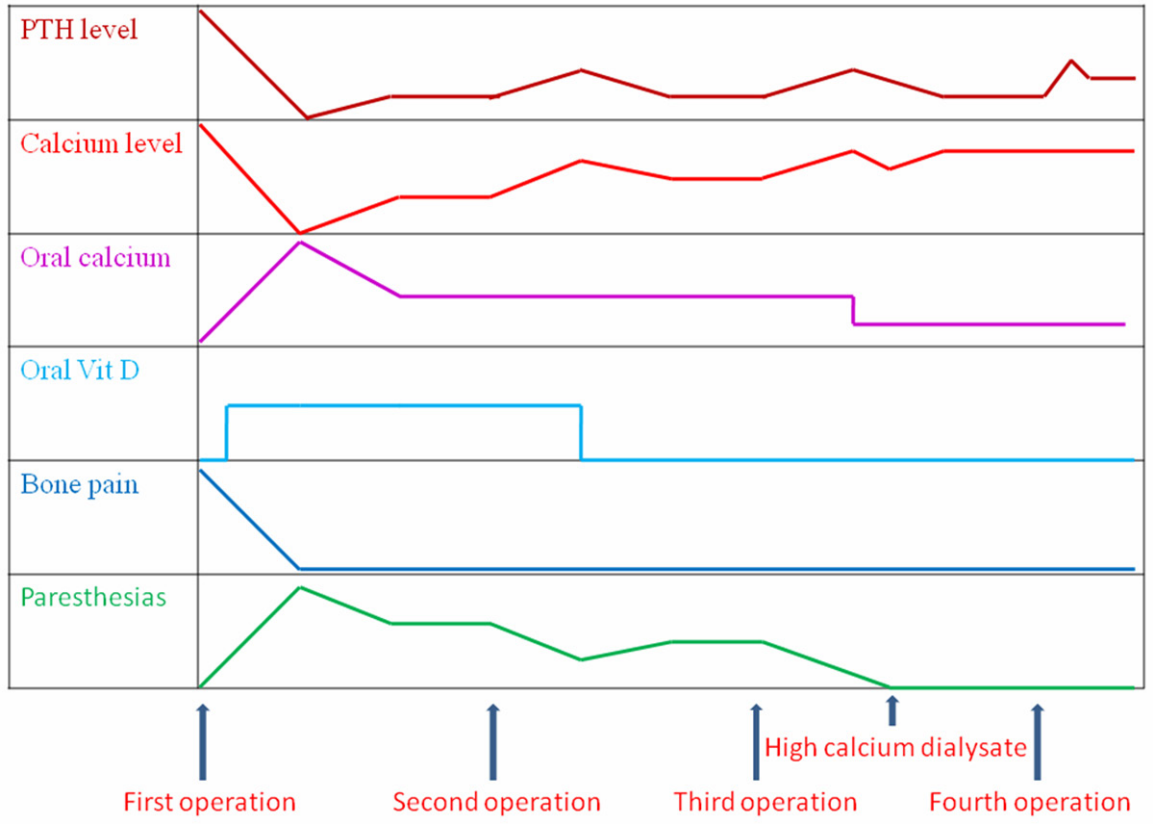

Figure 5. A diagrammatic sketch during the treatments and corresponding outcomes.

regular hemodialysis with a 1.75 mmol/L calcium dialysate every Tuesday, Thursday and Saturday. Symptoms of hypocalcemia such as paresthesias appeared every Monday. PTH was 15 ng/L and calcium was 1.52 mmol/L from non-grafted forearm 3 months after operation (**Figures 3** and **4**).

The second operation: replantation with cryopreserved parathyroid

The symptom was similar without alleviation 6 months after operation. He underwent the second operation: replantation with cryopreserved parathyroid into the same forearm in January 17, 2013. Three pieces of about 3 × 3 × 3 mm^3 cryopreserved parathyroid tissues were thawed, cut into 1 × 1 × 1 mm^3 pieces and injected into muscles of the forearm. PTH from grafted forearm increased to 420 ng/L one week later and decreased to 37 ng/L 1.5 months later (**Figures 3** and **4**). Meanwhile, he went on to receive oral calcium supplement without vitamin D. Paresthesias still appeared every Monday 3 months after the second transplantation.

The third operation: replantation with cryopreserved parathyroid

He underwent the third operation: replantation with cryopreserved parathyroid into the same forearm in April 17, 2013. PTH from grafted forearm increased to 118 ng/L 3 days later. Calcium concentration increased to 2.06 mmol/L, but decreased to 1.63 mmol/L 9 days later (**Figures 3** and **4**). The symptom got a little alleviation 9 days later, but aggravated again half a month later. 1.5 months after operation, he began to receive hemodialysis with 2.0 mmol/L calcium dialysate, then calcium concentration increased close to the normal level, and paresthesias disappeared. Then he decreased oral calcium supplement.

The fourth operation: replantation with cryopreserved parathyroid

The calcium concentration was close to normal by oral calcium supplement and hemodialysis with 2.0 mmol/L calcium dialysate. The PTH was still in the low limit of normal. Due to the chance of renal transplantation, he wanted to

Table 2. Definitions of parathyroid graft functional status according to different patients

	Non-functional	Partially functional	Fully functional
Dialysis patients	PTH ≤ 20	20 < PTH ≤ 50	50 < PTH ≤ 300
Non-dialysis patients	The same supplement	Decreased supplement	Without supplement
	PTH ratio ≤ 1.5	PTH ratio ≥ 1.5	PTH ratio ≥ 1.5

The supplement contains calcium and vitamin D administration.

receive the fourth operation to improve the calcium and PTH level without calcium supplement and dialysis after renal transplantation. He underwent replantation with cryopreserved parathyroid into muscles of the right forearm by open surgery in April 4, 2014. PTH from grafted forearm increased to 242 mmol/L, but from non-grafted forearm increased only a little. 20 days later, PTH still increased a little and calcium concentration was similar with preperation (**Figures 3** and **4**).

For better understanding, a diagrammatic sketch was made according to the process and outcome of the treatments (**Figure 5**).

Discussion

Herein, a patient with permanent hypoparathyroidism underwent four operations in our hospital. He was replanted with cryopreserved parathyroid in the last three operations. The duration of storage was 6 months, 9 months and 21 months between the first operation and the second, the third, the fourth operation respectively. The levels of PTH were higher from grafted forearm than from non-grafted forearm in the short term, but usually decreased 1 month later. The outcomes demonstrated that the cryopreserved parathyroid was active in the short term but reduced function in the long term. The outcome of the third operation was better than that of the second operation. One possible reason was that bone hungry syndrome was alleviated by calcium supplement 9 months after the first operation. The need of calcium is much less after the third operation than after the second operation. In addition, it may be attributed to the use of high calcium dialysate, which was recommended after the second operation, but his doctor of hemodialysis did not accept it. Because almost all uremic patients with hemodialysis are in high levels of calcium, some low calcium dialysates such as 1.25 mmol/L, 1.5 mmol/L and 1.75 mmol/L are used for them. Currently, a high calcium dialysate like 2.0 mmol/L needs to combine a low calcium dialysate with calcium solution before each use in our country. The less and inconvenient usage leads to the doctor not use a high calcium dialysate for him. Benefited from this case, a high calcium dialysate was efficient to increase calcium concentration and reduce symptoms of hypocalcemia. It might be an available treatment of persistent hypocalcemia and permanent hypoparathyroidism in dialysis patients. The clinical effect after the third operation satisfied us, but not the patient. Due to the chance of renal transplantation, he underwent the fourth operation to improve the calcium and PTH level to normal without calcium supplement and dialysis after renal transplantation. To date, there is no definite guideline about whether to transplant cryopreserved parathyroid for hypoparathyroidism but without hypocalcemia before renal transplantation. Based on the clinical effects of replantation, it could be considered as partial success in the short term. However, according to definitions for the function of cryopreserved parathyroid graft (**Table 2**) [2, 17], it should be defined as non-function for dialysis patients in the long term.

We searched for all papers related to replantation with cryopreserved parathyroid published in MEDLINE (National Library of Medicine, Bethesda, MD) from 1974 to 2014. The success rates of replantation with cryopreserved parathyroid vary from 20% to 100% [8-15, 17-19] (**Table 1**). Many factors affect the success of cryopreserved parathyroid graft. The function of cryopreserved parathyroid is the most important. Guerrero MA [21] found only 11 of 106 cryopreserved parathyroid specimens were viable. Only 1 of 92 specimens was viable when they were stored more than 24 months. On the contrary, 10 of 14 were viable when less than 24 months. A similar result was reported by Cohen MS [15]. Functional graft with cryopreserved parathyroid was only observed below 22 months of storage. Borot S

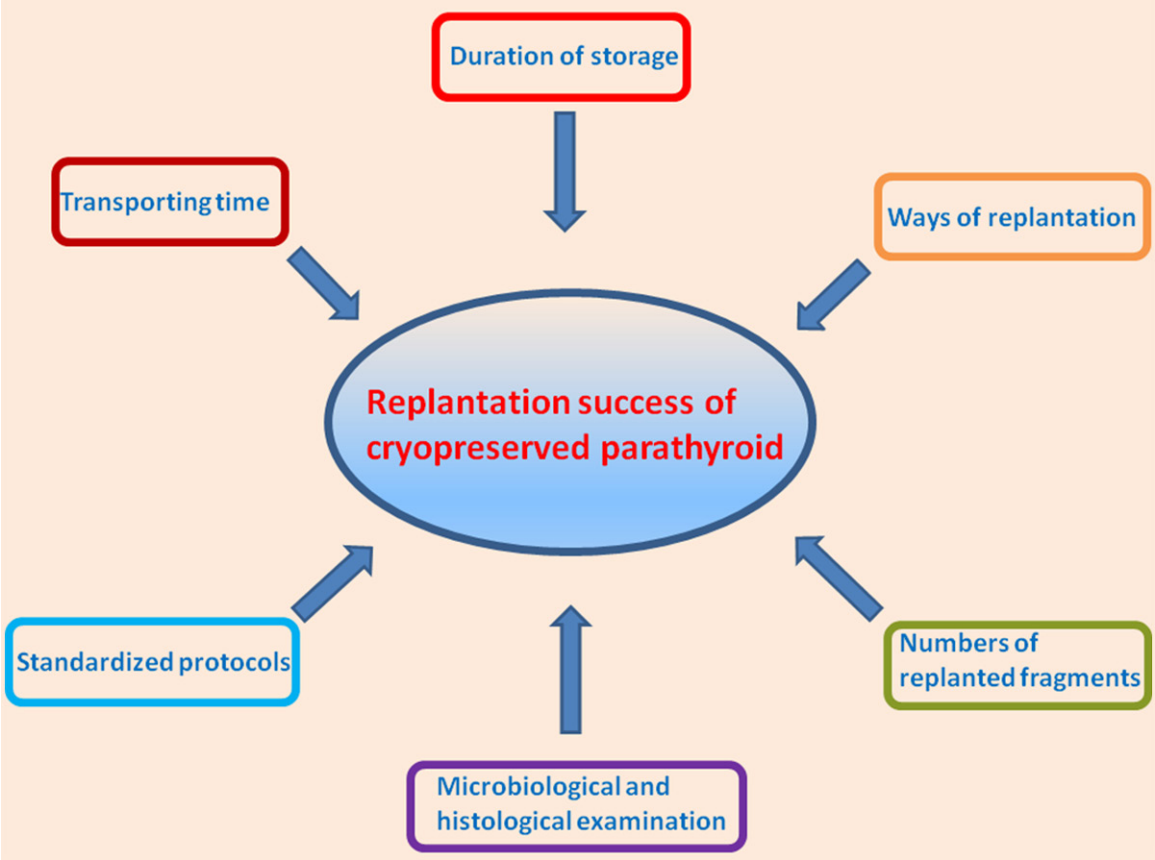

Figure 6. Factors related to the success of replantation with cryopreserved parathyroid.

reported only 4 of 20 grafts were functional from a retrospective multicenter study in France [17]. All of 4 grafts were cryopreserved less than 1 year. Therefore, it may be reasonable to disposal parathyroid specimen stored more than 24 months. However, Schneider R reported a 100% success rate of cryopreserved parathyroid in 15 permanent hypothyroidisms. 4 of them were cryopreserved than 2 years and the longest one was 86 months [18]. Several previous studies also displayed that the duration of cryopreservation was not closely related to the viability of cryopreserved cells in vitro [22-24] and the function in vivo [25]. From **Table 1**, the longest duration among the successful replantation was 125 months. Therefore, the duration of cryopreservation (6 months, 9 months and 21 months) might not be the main cause of failure in our case.

Recently, there are two ways of parathyroid transplantation by open surgery or by subcutaneous injection. Transplantation by open surgery is widely used, because it favors surgeons easy to re-explore hyper-functional grafted parathyroid by marks during operation. However, it leads to a long operation time, a big injury for grafted sites and possible general anesthesia. To avoid these shortcomings, transplantation by subcutaneous injection with local anesthesia may be a better choice. Two studies reported that the successful parathyroid transplantation by subcutaneous injection was similar with by open surgery, but with less complication and more convenience [26, 27]. Only one graft was hyper-functional, but it was easy to remove it [26]. Herein, the patient underwent four operations of parathyroid transplantation. The first and fourth were by open surgery, and the second and third were by subcutaneous injection. During the first operation, parathyroid transplantation was easy to mark without extra anesthesia simultaneously following total parathyroidectomy in case of re-exploration of hyperparathyroidism, which was also the reason of the fourth operation by open surgery. To save time of transporting thawed parathyroid from cryopreservation lab to opera-

tion room, parathyroid was implanted by subcutaneous injection during the second and third operation. What is the better way is still not definite. Currently, a proper way is favorable without loss of function, but with more convenience and less invasive injury. Therefore, the transplantation way might affect little on the failure of replantation with cryopreserved parathyroid in our case.

Some other factors also influence the success rate (**Figure 6**), such as the number of replanted fragments [28], microbiological and histological examination before replantation [18, 29]. Wagner PK suggested that 30-40 pieces of 1 mm^3 were sufficient in case of cryopreserved parathyroid with partial necrosis [22]. In our case, three pieces of about 3 × 3 × 3 mm^3 were thawed and replanted. The number was enough, but the activity was not examined before replantation. Schneider R only replanted tissues without necrosis or microbiological contamination by microbiological and histological examinations before replantation, and then they obtained a 100% success rate [18]. Stotler BA found 5 types of bacteria were present in 11 cryopreserved parathyroid tissues among 47 samples [29]. Thus, it is necessary to do microbiological examinations before replantation. We did not do microbiological and histological examinations before replantation due to lack of experiences of replantation. This might cause the failure in the long term in our case. Another reason might be the size of cryopreservation. Tissues were cryopreserved in the lab of reproductive medicine centre of our hospital without any cost. To reduce the costs, we cryopreserved three pieces of about 3 × 3 × 3 mm^3 in a tube. This was different from the standard size of 1 × 1 × 1 mm^3, which needs more places and more costs. Besides, a standardized freezing and thawing protocol [30], the transporting time between resection and freezing, thawing and transplantation [18] also affect the success of replantation with cryopreserved parathyroid.

Acknowledgements

The patient was informed and agreed to publish the pictures in any scientific journals.

Disclosure of conflict of interest

None.

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References

[1] Shoback D. Clinical practice. Hypoparathyroidism. N Engl J Med 2008; 359: 391-403.

[2] Guerrero MA. Cryopreservation of parathyroid glands. Int J Endocrinol 2010; 2010: 829540.

[3] Winer KK, Ko CW, Reynolds JC, Dowdy K, Keil M, Peterson D, Gerber LH, McGarvey C and Cutler GB. Long-term treatment of hypoparathyroidism: a randomized controlled study comparing parathyroid hormone- (1-34) versus calcitriol and calcium. J Clin Endocrinol Metab 2003; 88: 4214-4220.

[4] Rubin MR, Sliney J, McMahon DJ, Silverberg SJ and Bilezikian JP. Therapy of hypoparathyroidism with intact parathyroid hormone. Osteoporos Int 2010; 21: 1927-1934.

[5] Sikjaer T, Rejnmark L, Rolighed L, Heickendorff L, Mosekilde L; Hypoparathyroid Study Group. The effect of adding PTH (1-84) to conventional treatment of hypoparathyroidism: a randomized, placebo-controlled study. J Bone Miner Res 2011; 26: 2358-2370.

[6] Cusano NE, Rubin MR, McMahon DJ, Zhang C, Ives R, Tulley A, Sliney J, Cremers SC and Bilezikian JP. Therapy of hypoparathyroidism with PTH (1-84): a prospective four-year investigation of efficacy and safety. J Clin Endocrinol Metab 2013; 98: 137-144.

[7] Wells SA and Christiansen C. The transplanted parathyroid gland: evaluation of cryopreservation and other environmental factors which affect its function. Surgery 1974; 75: 49-55.

[8] Brennan MF, Brown EM, Spiegel AM, Marx SJ, Doppman JL, Jones DC and Aurbach GD. Autotransplantation of cryopreserved parathyroid tissue in man. Ann Surg 1979; 189: 139-142.

[9] Wells SA Jr, Farndon JR, Dale JK, Leight GS and Dilley WG. Long-term evaluation of patients with primary parathyroid hyperplasia managed by total parathyroidectomy and heterotopic autotransplantation. Ann Surg 1980; 192: 451-458.

[10] Saxe AW, Spiegel AM, Marx SJ and Brennan MF. Deferred parathyroid autografts with cryopreserved tissue after reoperative parathyroid surgery. Arch Surg 1982; 117: 538-543.

[11] Wagner PK, Seesko HG and Rothmund M. Replantation of cryopreserved human parathyroid tissue. World J Surg 1991; 15: 751-755.

[12] Herrera M, Grant C, van Heerden JA and Fitzpatrick LA. Parathyroid autotransplantation. Arch Surg 1992; 127: 825-829; discussion 829-830.
[13] Caccitolo JA, Farley DR, van Heerden JA, Grant CS, Thompson GB and Sterioff S. The current role of parathyroid cryopreservation and autotransplantation in parathyroid surgery: an institutional experience. Surgery 1997; 122: 1062-1067.
[14] Feldman AL, Sharaf RN, Skarulis MC, Bartlett DL, Libutti SK, Weinstein LS, Marx SJ, Norton JA, Fraker DL and Alexander HR. Results of heterotopic parathyroid autotransplantation: a 13-year experience. Surgery 1999; 126: 1042-1048.
[15] Cohen MS, Dilley WG, Wells SA, Moley JF, Doherty GM, Sicard GA, Skinner MA, Norton JA, DeBenedetti MK and Lairmore TC. Long-term functionality of cryopreserved parathyroid autografts: a 13-year prospective analysis. Surgery 2005; 138: 1033-1040; discussion 1040-1031.
[16] de Menezes Montenegro FL, Custodio MR, Arap SS, Reis LM, Sonohara S, Castro IV, Jorgetti V, Cordeiro AC and Ferraz AR. Successful implant of long-term cryopreserved parathyroid glands after total parathyroidectomy. Head Neck 2007; 29: 296-300.
[17] Borot S, Lapierre V, Carnaille B, Goudet P and Penfornis A. Results of cryopreserved parathyroid autografts: a retrospective multicenter study. Surgery 2010; 147: 529-535.
[18] Schneider R, Ramaswamy A, Slater EP, Bartsch DK and Schlosser K. Cryopreservation of parathyroid tissue after parathyroid surgery for renal hyperparathyroidism: does it really make sense? World J Surg 2012; 36: 2598-2604.
[19] Shepet K, Alhefdhi A, Usedom R, Sippel R and Chen H. Parathyroid cryopreservation after parathyroidectomy: a worthwhile practice? Ann Surg Oncol 2013; 20: 2256-2260.
[20] Leite AK, Junior CP, Arap SS, Massoni L, Lourenço DM, Brandão LG and Montenegro FL. Successful parathyroid tissue autograft after 3 years of cryopreservation: a case report. Arq Bras Endocrinol Metabol 2014; 58: 313-316.
[21] Guerrero MA, Evans DB, Lee JE, Bao R, Bereket A, Gantela S, Griffin GD and Perrier ND. Viability of cryopreserved parathyroid tissue: when is continued storage versus disposal indicated? World J Surg 2008; 32: 836-839.
[22] Wagner PK, Rumpelt HJ, Krause U and Rothmund M. The effect of cryopreservation on hormone secretion in vitro and morphology of human parathyroid tissue. Surgery 1986; 99: 257-264.
[23] Herrera MF, Grant CS, van Heerden JA, Jacobsen D, Weaver A and Fitzpatrick LA. The effect of cryopreservation on cell viability and hormone secretion in human parathyroid tissue. Surgery 1992; 112: 1096-1101; discussion 1101-1092.
[24] McHenry CR, Stenger DB and Calandro NK. The effect of cryopreservation on parathyroid cell viability and function. Am J Surg 1997; 174: 481-484.
[25] Rothmund M and Wagner PK. Assessment of parathyroid graft function after autotransplantation of fresh and cryopreserved tissue. World J Surg 1984; 8: 527-533.
[26] Echenique-Elizondo M, Amondarain JA, Vidaur F, Olalla C, Aribe F, Garrido A, Molina J and Rodrigo MT. Parathyroid subcutaneous presternal transplantation after parathyroidectomy for renal hyperparathyroidism. Long-term graft function. World J Surg 2007; 31: 1403-1409.
[27] Yoon JH, Nam KH, Chang HS, Chung WY and Park CS. Total parathyroidectomy and autotransplantation by the subcutaneous injection technique in secondary hyperparathyroidism. Surg Today 2006; 36: 304-307.
[28] Wagner PK and Rothmund M. [Replantation of autologous cryopreserved parathyroid tissue in permanent postoperative hypoparathyroidism]. Dtsch Med Wochenschr 1987; 112: 1160-1162.
[29] Stotler BA, Reich-Slotky R, Schwartz J, Inabnet WB, Lee J, Wu F, Della-Latta P and Jhang JS. Quality monitoring of microbial contamination of cryopreserved parathyroid tissue. Cell Tissue Bank 2011; 12: 111-116.
[30] Agarwal A, Waghray A, Gupta S, Sharma R and Milas M. Cryopreservation of parathyroid tissue: an illustrated technique using the cleveland clinic protocol. J Am Coll Surg 2013; 216: e1-9.