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Gestational trophoblastic diseases: 4. Presentation with persistent low positive human chorionic gonadotropin test results

Laurence A. Cole [a,*], Sarah A. Khanlian [a], Almareena Giddings [a], Stephen A. Butler [a], Carolyn Y. Muller [b], Charles Hammond [c], Ernest Kohorn [d]

[a] USA hCG Reference Service, Department of Obstetrics and Gynecology, MSC10 5580 1, University of New Mexico, Albuquerque, NM 87131-0001, USA
[b] Division of Gynecologic Oncology, University of New Mexico, Albuquerque, NM 87131, USA
[c] Southeastern Regional Trophoblastic Center, Department of Obstetrics and Gynecology, Duke University Medical Center, Durham, NC 27710, USA
[d] Yale Trophoblast Center, Department of Obstetrics and Gynecology, Yale University, New Haven, CT 06520, USA

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Abstract

Objectives. A high proportion of women with persistent low levels of hCG, in the absence of pregnancy or any evidence of tumor, have received chemotherapy and hysterectomy for assumed malignancy. Such chemotherapy and surgery were ineffective and unwarranted. This study identifies the causes of persistent low level of hCG and provides guidelines for the management of these patients, preventing unnecessary treatment in the future.

Methods. The USA hCG Reference Service has consulted on 170 women with low levels of hCG persisting for 3 months or longer. Serum total hCG was measured in the Diagnostic Products Corporation (DPC) Immulite assay and hyperglycosylated hCG in the Nichols Advantage test.

Results. Among these 170 patients, the average persistent hCG result was 102 ± 152 mIU/ml, with a range of 6.1–900 mIU/ml. Thirteen (7.6%) of the 170 patients had true malignancy, 5 had placental site trophoblastic tumor, 3 had other gestational trophoblastic neoplasms (GTN), and 5 had non-trophoblastic malignancies. The remaining 157 patients had false-positive hCG, quiescent gestational trophoblastic disease (quiescent GTD), or pituitary hCG (hCG of pituitary origin).

Of 71 patients with false-positive hCG, 47 patients received chemotherapy and 9 had surgery that had no effect on the level of hCG. Five of these patients with false-positive hCG were being monitored for hydatidiform mole or GTN. The majority of these cases were first investigated following an incidental pregnancy test.

Of 69 patients who had quiescent GTD, 41 received chemotherapy and 9 underwent hysterectomy. All these therapies were unnecessary and ineffective. While 21 patients with quiescent GTD followed incidental pregnancy tests, the majority were discovered while monitoring patients after treatment for hydatidiform mole or GTN/choriocarcinoma (n = 48). Seventeen cases of pituitary hCG were found among those women who were peri- or post-menopause. Two patients also received chemotherapy for assumed malignancy which was not present.

Conclusion. Clinicians frequently assume that an elevated hCG implies that a patient is pregnant or has GTD or recurrent GTN, even when apart from the pregnancy test, no clinical evidence was found to support such a diagnosis. In most of these cases of persistent low hCG etiologies, all therapies were found unnecessary and ineffective. Guidelines are proposed for managing these patients. It is essential to demonstrate a malignancy clinically and with readily available biochemical tests before initiating therapy. This applies whether the patient is identified by an incidental pregnancy test or is actively being monitored for gestational trophoblastic disease.

Keywords: Hyperglycosylated hCG; hCG-H; hCG human chorionic; Gonadotropin; Chriocarcinoma; Cytokine; Hyperglycosylated hCG

Introduction

The USA hCG Reference Service has been consulted in an increasing proportion of patients identified with persistent low levels of hCG [1–5]. These present in two clear sets of circumstances. In the first group, the patient has a recent

* Corresponding author. Fax: +1 505 272 3576.
E-mail address: larry@hcglab.com (L.A. Cole).

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history of hydatidiform mole or choriocarcinoma with confirming histology, or gestational trophoblastic neoplasm (GTN), based on history and elevated hCG levels but without histology. Following successful uterine evacuation or chemotherapy, persistent low levels of serum hCG are detected, persisting from 3 months (minimum time for us to call it persistent) to greater than 10 years [3,6]. Three etiologies have been demonstrated for this kind of finding: 1. false-positive hCG due to a poorly designed hCG test, 2. quiescent gestational trophoblastic disease (GTD), and 3. pituitary hCG [1–6]. In the second group, a patient had an incidental pregnancy test as part of an obstetrics-gynecology evaluation, prior to imaging or minor surgery. Pregnancy is assumed and then excluded by ultrasound and subsequent dilation and curettage. In these patients, a positive serum hCG persists with minimal variations from 3 months to greater than 10 years. Current practice dictates treatment of those with elevated hCG, GTN is commonly inferred, and in a high proportion of patients single agent chemotherapy is commenced or hysterectomy is performed [1–6]. A few patients are in fact found to have active choriocarcinoma/GTN, placental site trophoblastic tumor (PSTT), or non-trophoblastic malignancy, in which case therapy is needed. We most commonly find a more benign source, false-positive hCG test, quiescent GTD, or pituitary hCG. In such cases, anti-cancer treatment is ineffective [1–7].

Here, we present the USA hCG Reference Service experience consulting with 170 patients who were found to have persistent low levels of hCG. Based on this extensive experience, guidelines are presented on managing these patients and on avoiding inappropriate and ineffective therapy.

Methods

Patients

The nomenclature used throughout this presentation is that of FIGO and the SGO. GTD is a general term for all trophoblastic diseases, while GTN applies to trophoblastic malignancies that are histologically choriocarcinoma or trophoblastic neoplasms without histologic verification. PSTT is reported separately.

Subjects were all women referred to and supplying serum samples to the USA hCG Reference Service at Yale University and the University of New Mexico, USA, between January 1998 and June 2004. This article concentrates on patients with low level hCG, persisting for 3 months or longer. These patients had either false-positive hCG results or quiescent GTD or were found to have pituitary hCG or were those in which persistent low levels of hCG are due to active malignancy. Seventy one patients had false-positive hCG. This was based on meeting all 3 of 3 criterion of the USA hCG Reference Service [1–5]. These are: 1. the presence of hCG immunoreactivity in serum but not urine; 2. varying hCG results (more than 5-fold) or negative results in 3 or more hCG tests; 3. the suppression of a positive finding by a heterophilic antibody blocking agent. The 17 pituitary hCG patients were peri- or post-menopausal women with low levels of hCG, <25 mIU/ml, in whom false-positive hCG, non-trophoblastic malignancy, and choriocarcinoma/GTN or placental site trophoblastic malignancy had been excluded. The 69 patients with quiescent GTD and the 13 patients presenting with persistent low hCG results that were later diagnosed as GTN, PSTT, or having non-trophoblastic malignancy are the patients described in the preceding publications [6,7]. Evaluation of the databases of digitized USA hCG Reference Service data and examination of patient records were all approved by the University of New Mexico Human Research Review Committee (protocols 99-349 and 02-548).

Laboratory tests

All laboratory testing was performed in the USA hCG Reference Service laboratories. This laboratory is certified by the Department of Heath and Human Services for performing clinical tests for patient records (CLIA certification 32D0972561). The consistency of laboratory tests is monitored by the College of American Pathologists (CAP certification 7176750-01).

Serum samples were received frozen then thawed and tested immediately. All basic testing involved automated assays, using pre-formulated reagent packs. Serum and urine total hCG were measured using the robotic chemiluminescence DPC Immulite hCG test (DPC Inc., Los Angeles, CA). This assay detects hCG, hCG-H, and free β-subunit on an equal molar basis. When the concentration of pure hCG, hCG-H, and free β-subunit was determined in molar units (nmol) by absorbance at 278 nm, near-identical results were observed (in mIU/ml) in the DPC Immulite hCG test (H-hCG result 99% and free β-subunit result 100% of hCG standard concentration) [2,3,6].

It should be noted that, while the total hCG, hCG-H, and hCG free β-subunit assay are all commercially available and are all FDA-approved tests, they are only approved for pregnancy applications. Gestational trophoblastic diseases can be considered as pregnancy or gestation-related applications, but these cancer-related applications should be considered as "off-label" applications. We have carefully evaluated all 3 tests and demonstrated their particular suitability and accuracy, compared with other commercial hCG tests, for gestational trophoblastic disease applications [1–6,10].

Data analysis

In June 2005, all accrued test results, 1998–2005, dates, ages, diagnoses, antecedent gestation data, and pertinent treatment histories were digitized by entry into Microsoft Excel 2003 spreadsheet (Microsoft Inc., Redmond WA). Basic mean, range and standard deviation statistics, and *t* statistics were determined in the Excel 2003 spreadsheet. Data groups were ranked, and non-parametric centiles were determined, and detection rates were calculated at corresponding false-positive rates.

Results

Overview of persistent low hCG patients

The USA hCG Reference Service has consulted on 170 patients with persistent low hCG results, in each case, there was absence of any type of tumor, physical evidence of malignancy, or intra- or extra-uterine pregnancy. As defined, persistent low levels of hCG (<1000 mIU/ml) are those that plateau while maintaining some variance in repeated measurements. This is week-to-week variations in low levels due to changes in body hydration, immune system, menstrual hormone levels, and other not readily explained factors that make small day to day variation in false-positive, quiescent, and pituitary hCG results with no consistent inclining or declining trend.

Retrospective studies

Of the 170 patients referred with persistent low level hCG, 71 (42%) were found to be due to false-positive hCG test results, 69 (41%) had quiescent GTD, 17 (10%) had pituitary hCG production, and 13 (7.6%) had active malignancy, either GTN, PSTT, or non-trophoblastic malignancy (Table 1). Overall, approximately one third of cases occurred in those with history of GTD/GTN ($n = 59$) and two thirds in those with no history of GTD/GTN ($n = 111$). In the former patients,

Table 1
The USA hCG Reference Service experience with 170 cases with persistent low levels of hCG persisting for 3 months or longer, with no evidence of pregnancy or imaging evidence of tumor

Diagnosis GTN/choriocarcinoma	
History of cases	
Discovered by incidental pregnancy test	3 of 3
Antecedent event, term pregnancy	3 of 3
Effective surgery/chemotherapy	3 of 3
Ineffective or unnecessary surgery/chemotherapy	0 of 3
Persistent hCG test results at presentation	
Mean ± standard deviation	1769 ± 3048 mIU/ml [a]
Range	9.1–5290 mIU/ml
Diagnosis PSTT	
History of cases	
Discovered by incidental pregnancy test	2 of 5
Discovered post-evacuation of hydatidiform mole	3 of 5
Effective surgery/chemotherapy	5 of 5
Ineffective or unnecessary surgery/chemotherapy	0 of 5
Persistent hCG test results at presentation	
Mean ± standard deviation	63 ± 94 mIU/ml[a]
Range	8.5–231 mIU/ml
Diagnosis of non-trophoblastic malignancy	
History of cases	
Discovered by incidental pregnancy test	5 of 5
Germ cell malignancy	4 of 5
Other malignancy	1 of 5 cases
Effective surgery/chemotherapy	5 of 5 cases
Ineffective or unnecessary surgery/chemotherapy	0 of 5 cases
Persistent hCG test results at presentation	
Mean ± standard deviation	38 ± 68 mIU/ml[a]
Range	2.9–160 mIU/ml
Diagnosis quiescent GTD	
History of cases	
Discovered by incidental pregnancy test	21 of 69
Discovered post-evacuation of hydatidiform mole	33 of 69
Discovered after treatment of choriocarcinoma/GTN	15 of 69
Cases receiving unnecessary therapy	42 of 69 (41 chemotherapy, 7 hysterectomy)
Effective surgery/chemotherapy	0 of 69
Duration of persistent hCG results before referral	3 months–16 years
Age (mean ± standard deviation)	34 ± 8.5
hCG and related test results at presentation	
Mean ± standard deviation	55 ± 103 mIU/ml[a]
Range	0.5–161 mIU/ml
Diagnosis false-positive hCG test	
History of cases	
Discovered by incidental pregnancy test	66 of 71
Discovered post-evacuation of hydatidiform mole	3 of 71
Discovered after treatment of choriocarcinoma/GTN	2 of 71
Cases receiving unnecessary therapy	47 of 71 (46 chemotherapy, 9 hysterectomy)
Diagnosis false-positive hCG test	
History of cases	
Effective surgery/chemotherapy	0 of 71
Duration of persistent hCG results before referral	3 months–2 years
Age (mean ± standard deviation)	34 ± 6.4
hCG and related test results at presentation	
Mean ± standard deviation	102 ± 152 mIU/ml[a]
Range	6.1–900 mIU/ml
Diagnosis pituitary hCG	
History of cases	
Discovered by incidental pregnancy test	11 of 17
Discovered post-evacuation of hydatidiform mole	5 of 17
Discovered after treatment of choriocarcinoma/GTN	1 of 17
Cases receiving unnecessary therapy	2 of 17 (2 chemotherapy)
Effective surgery/chemotherapy	0 of 17
Duration of persistent hCG results before referral	
Suppressed with hormone replacement therapy	11 of 11 (feedback on 11)
Age (mean ± standard deviation)	51 ± 8.8[b]
hCG and related test results at presentation	
Mean ± standard deviation	7.6 ± 4.6[c]
Range	1.2–19

The 71 cases diagnosed as having false-positive hCG were based on multiple observations by the USA hCG Reference Service. These are: 1. the presence of hCG immunoreactivity in serum but not urine; 2. varying hCG results (more than 5-fold) or negative results in 3 or more hCG tests; 3. the suppression of result by a heterophilic antibody blocking agent. The 17 pituitary hCG cases were defined as those peri- or post-menopausal women with low levels of hCG <25 mIU/ml, having excluded false-positive hCG, non-trophoblastic malignancy and choriocarcinoma/GTN or placental site trophoblastic malignancy. The 69 cases with quiescent GTD and the 11 cases with PSTT are those described in preceding publications [6,7]. All hCG values are those determined at the USA hCG Reference Service.

[a] While the average hCG for the 3 GTN/choriocarcinoma cases is significantly higher than that in false-positive hCG and quiescent GTD, it is due to the wide range of the cases, 5290, 9.2, and 9.1 mIU/ml. No significant difference was observed in the ranges of hCG results in PSTT, non-trophoblastic malignancy, quiescent GTD, and false-positive hCG outcome cases.

persistent low levels of hCG were found during follow-up of hydatidiform mole and GTN/choriocarcinoma patients. This occurred either as a residual low level of hCG after evacuation of hydatidiform mole, after primary therapy of GTN, or after hCG reappeared in the months after hCG became undetectable. In the latter or majority of patients, persistent low levels of hCG were found in an incidental home pregnancy test or from an incidental serum hCG test prior to minor surgery, radioimaging, or an infertility or gynecologic examination.

As shown in Table 1, neither the level of the persistent low hCG nor the age of the patient provided significant information regarding the origin of persistent low levels of hCG ($P > 0.05$). The exception is pituitary hCG, which can be distinguished from quiescent GTD ($P < 0.0000001$) or false-positive hCG ($P < 0.0000001$) cases by being produced in significantly older females, that are either menopausal or peri-menopausal.

Table 2
The 71 false-positive hCG patients that consulted with the USA HCG Reference Service

Physician's diagnosis according to records	hCG tests	hCG mIU/ml [a]	Previous history (prior 6 months)	Ineffective chemotherapy or surgery
GTN	Abbott [b]	900	None	Mtx
GTN	Abbott × 3 [c]	600	None	Mtx, ActD, Hys, EMA-CO, Tho
GTN	Abbott	500	None	Mtx
Choriocarcinoma	Abbott	467	None	Mtx, ActD, EMA-CO, Hys
GTN	Abbott	350	Parturition	Mtx
GTN	Abbott	275	None	
None	Abbott	271	None	
GTN	Abbott	218	None	Mtx
GTN	Abbott	205	None	
GTN	Abbott	200	None	Mtx
None	Abbott	174	None	
GTN	Abbott	160	None	Mtx, ActD
GTN	Beckman	153	None	Mtx, EMA-CO
None	Abbott	151	None	Mtx
GTN	Abbott	143	None	
GTN	Abbott	142	Miscarriage	Mtx, ActD, Hys
GTN	Abbott	139	None	Mtx
Ectopic pregnancy	Abbott	122	None	Mtx
None	Bayer	117	None	Mtx
GTN	Ortho	100	Miscarriage	Hys
GTN	Abbott	97	None	Mtx
GTN	Abbott	83	None	Mtx
GTN	Abbott	81	None	Mtx
GTN	Abbott	81	None	Mtx, ActD, Hys, SalOoph
Choriocarcinoma	Abbott	80	Choriocarcinoma	Etop
GTN	Abbott	80	None	Mtx, EMA-CO
Choriocarcinoma	Abbott	78	None	Mtx, Hys, EMA-CO
GTN	Beckman	78	None	
GTN	Abbott	74	None	
GTN	Abbott	60	None	
GTN	Abbott	60	None	
GTN	Abbott	57	None	Mtx
None	Ortho	55	Parturition	
Persistent mole	Abbott	53	Complete Mole	
None	Bayer	50	None	
Choriocarcinoma	Abbott	47	None	Mtx, ActD, EMA-CO, Hys
GTN	Abbott	42	None	Mtx, ActD
GTN	Ortho	41	Complete Mole	Mtx, ActD
GTN	Abbott	40	None	Mtx
GTN	Abbott	37	Miscarriage	Mtx, Mtx
Persistent mole	Abbott	35	Complete Mole	Mtx, Hys
GTN	Abbott	33	None	Mtx
GTN	Bayer	32	None	Mtx
GTN	Bayer	30	None	Mtx
GTN	Abbott	26	None	Mtx
None	Beckman	25	None	Mtx
GTN	Abbott	24	None	Hys
GTN	Abbott	23	None	Mtx
GTN	Dade	23	Miscarriage	Mtx
GTN	Abbott	22	None	Mtx, ActD
Persistent mole	Abbott	21	Partial Mole	Mtx, AcD
GTN	Bayer	20	None	Mtx
None	Bayer	19	None	
GTN	Abbott	18	None	Mtx
GTN	Abbott	17	None	Mtx, Hys
None	Abbott	16	None	
GTN	Abbott	14	GTN	
None	Bayer	13	Miscarriage	
None	Abbott	12	None	Hys, SalOoph
None	Bayer	12	None	
Residual trophoblast tissue	Abbott	12	Miscarriage	
GTN	Bayer	12	None	Mtx
GTN	Abbott	11	None	Mtx
GTN	Bayer	11	None	Mtx
None	Abbott	10	None	Mtx
GTN	Ortho	9	None	
GTN	Bayer	9	GTN	Mtx
None	Bayer	8.5	None	
Persistent mole	Abbott	8	Complete Mole	Mtx, ActD, EMA-CO, Hys
GTN	Tosoh	7.1	Choriocarcinoma	
None	Bayer	6.1	None	

All hCG data are hCG Reference Service serum results. Therapy abbreviations — methotrexate (Mtx); actinomycin D (ActD); etoposide + methotrexate + actinomycin D, alternating with cyclophosphamide + vincristine (EMA-CO); hysterectomy (Hys); salpingo-oophorectomy (SalOoph); thoracotomy (Tho). Abbreviations for hCG tests — Abbott AxSym test (Abbott); Beckman Access (Beckman); three analogous Bayer tests, Bayer ACS180, Bayer ADVIA Centaur, and Bayer ACS180 (Bayer); Ortho Vitros (Ortho); Dade Dimension RXL (Dade); Tosoh A1A (Tosoh).

[a] hCG results at the time of or immediately prior to referral to the USA hCG Reference Service.

[b] Of the 71 patients, 49 were managed by their laboratory using the Abbott AxSym, 13 using a Bayer analogous assay, 4 using the Ortho Vitros, 3 using the Beckman Access, and 1 each using the Dade Dimension RXL and the Tosoh AIA quantitative automated total hCG test.

[c] Prior to hysterectomy, hCG was tested at 3 independent laboratories, all gave the same false-positive hCG result since all used the Abbott AxSym test.

Pituitary hCG patients also have significantly lower hCG results than those with quiescent GTD or false-positive hCG test, $P < 0.0000001$ and $P < 0.0000001$.

False-positive hCG

Table 2 summarizes the experience with the 71 patients shown to have false-positive hCG tests. Among these women, the average persistent hCG result was 102 ± 152 mIU/ml, with a range of 6.1–900 mIU/ml. The majority of false-positive hCG results were from women having incidental pregnancy tests. Forty seven patients received chemotherapy, and 12 had surgery for what was later shown to be an hCG test problem. Five patients were being monitored after hydatidiform mole or GTN. Of the 71 patients, 49 (69% of cases) were managed by their center's laboratory using the Abbott AxSym, 13 (18% of cases) using a Bayer analogous assays, 4 (6% of cases) using the Ortho Vitros, 3 (4% of cases) using the Beckman Access, and 1 (1% of cases) each using the Dade Dimension RXL and the Tosoh AIA quantitative automated total hCG test. It is noteworthy that a

high proportion of cases observed in the past 2 years have been using the Bayer analogous assays and fewer using the Abbott AxSym test [1,2,4,5].

Quiescent GTD

In the second paper in this series, Table 2 [6] summarizes the 69 patients with quiescent GTD. Among these patients, the average hCG was 55 ± 103 mIU/ml, with a range of 0.5–161 mIU/ml. Twenty one patients with quiescent GTD were found by incidental pregnancy tests in patients with no history of GTD. The majority, however, were discovered while monitoring patients after evacuation of hydatidiform mole (n = 33) or after treatment of GTN/choriocarcinoma (n = 15). Forty one patients with quiescent GTD received chemotherapy and 9 had a hysterectomy. All therapies failed to suppress the hCG production in all patients.

Pituitary hCG

Table 3 summarizes the experience with 17 patients who had pituitary hCG. The average hCG was 7.6 ± 4.6 mIU/ml with a range of 1.2–19 mIU/ml. In all these patients found to have pituitary hCG, the USA hCG Reference Service suggested the use of steroid hormone replacement or oral contraceptive therapy to suppress production of hCG and confirm its pituitary origin. In 5 patients, repeat hCG testing, after 3 weeks therapy, was performed by the Service. In all 5 patients, steroids suppressed hCG production. In a further 6 patients, the Service received feedback that hormone replacement therapy suppressed hCG. Therefore, in all 11 patients, hCG was proven to be of pituitary origin. In 2 patients with a history of complete mole in the preceding 3 years, GTN was assumed and methotrexate chemotherapy given. This had no effect on suppressing hCG production.

Case reports

Three cases are presented to illustrate the ineffective and seemingly unnecessary treatment given to women with non-threatening persistent low levels of hCG.

Case 1, a representative example of quiescent GTD

The patient had persistent low levels of hCG of approximately 40 mIU/ml lasting for over 2 years. During this period, she received single agent methotrexate followed by actinomycin D. With no resolution of serum hCG, she was treated with EMA-CO (etoposide, methotrexate, actinomycin D alternating with cyclophosphamide and vincristin) combination chemotherapy followed by a hysterectomy. The hCG remained elevated. She was then referred to our Service, where hCG levels and history were evaluated. The absence of hCG-H indicated that no active metastatic disease was present [1–6]. The indicated diagnosis was quiescent GTD, and then all therapy was halted. We were informed that 1 year after the referral the persistent low hCG was spontaneously resolved (Table 4).

Table 3
Seventeen peri-menopause or menopause patients producing pituitary hCG

Age	hCG mIU/ml	Physician's indicated diagnosis according to records	Ineffective therapy	Previous history (prior 3 years)	Suppression by HRT[a]
35	19	GTN	Mtx	Complete mole	Confirmed
59	16	False-Positive hCG			Confirmed
48	11.4	None		Partial mole	Confirmed
57	11	None			
54	10.2	GTN		GTN	Confirmed
52	9	GTN		Complete mole	Confirmed
57	7.9	Non-trophoblastic Malignancy		Breast malignancy	Confirmed
51	7.4	False-Positive hCG			
56	7.3	Non-trophoblastic Malignancy			Confirmed
53	7.2	None			
53	5.9	GTN		Complete mole	Confirmed
39	5.8	Non-trophoblastic Malignancy		Post-oophorectomy	
47	5.3	None			Confirmed
57	3.7	None			Confirmed
69	3.5	None			
51	3.3	GTN	Mtx	Complete mole	Confirmed
49	2.1	None			
37	1.2	False-Positive hCG		Post-oophorectomy	

Two patients received methotrexate (Mtx) chemotherapy. Pituitary hCG patients were defined as those peri- or post-menopausal with low levels of hCG <25 mIU/ml, having excluded false-positive hCG, non-trophoblastic malignancy, and choriocarcinoma/GTN or placental site trophoblastic malignancy. All hCG data are hCG Reference Service results.

[a] USA hCG Reference Service deduced pituitary hCG and suggested the use of a high dose hormone replacement therapy to suppress production and confirm pituitary origin. In 5 patients repeat hCG testing, after 3 week therapy, was performed by the Reference Service. In all 5 patients steroids suppressed hCG production. In a further 6 patients the Reference Service received feedback that hormone replacement therapy suppressed hCG.

Case 2, quiescent GTD leading to active GTN/choriocarcinoma and death

An incidental positive pregnancy test led to an ultrasound and subsequently to a laparoscopic salpingectomy and excision of an ectopic mass. The pathology concluded this was choriocarcinoma, but review of the descriptive findings (necrotic, primarily large syncytiotrophoblast cells) would be more supportive of an ectopic hydatidiform mole or an ectopic quiescent GTD rather than choriocarcinoma [9]. The final diagnosis was tubal non-metastatic choriocarcinoma (assumingly, the same as quiescent GTD). After this surgical removal, hCG results cleared over 2 months as would normally occur following an ectopic pregnancy. A residual level of 15 mIU/ml hCG was detected and persisted in this range for 3 years. During this time, this patient received an extensive array of different chemotherapy regimens including

Table 4
Representative case of quiescent GTD persisting for over 2 years, treated without success by multiple chemotherapy regimens and hysterectomy

Month	hCG mIU/ml	Progress and comments
0	44	Incidental pregnancy test, confirmed with serum test
0	40	D and C reveal no evidence of pregnancy
0	38	Laparoscopy reveals no ectopic pregnancy
3	52	
3	38	MRI/CT of head, chest, and pelvis is unremarkable
8	40	
9	35	Start 4 regimens methotrexate chemotherapy
10	15	Start 4 regimens actinomycin D chemotherapy
21	60	CT scan of pelvis indicates uterine irregularities
22	60	Hysterectomy, pathology unremarkable
23	13	
23	20	Start 3 EMA-CO (etoposide/methotrexate/actinomycin D, alternating with cyclophosphamide and vincristine) diweekly regimens
26	20	
27	24	Referral to hCG Ref. Service, quiescent GTD concluded, all treatment stopped
40	0	Reported to the USA hCG Ref. Service, quiescent GTD spontaneously resolved

Patient age 41, para 2, gravida 3, case started with incidental pregnancy test 4 months after spontaneous abortion. All hCG data are from patients records, or testing institution, except as indicated.

methotrexate, EMA-CO, MAC (methotrexate, actinomycin D and cyclophosphamide), and TP (taxol and cisplatin) with no steady change in the hCG results. She then underwent hysterectomy and bilateral salpingo-oophorectomy. This also failed to suppress the hCG. She was then given regimens of VIP (VP-16, ifosphamide, cisplatin), Xeloda (capecitabine), and regimens of hydroxyurea. During these 3 years, the Reference Service was twice consulted on 2 occasions, once at the request of the patient and once at the request of an independent consultant. The absence of hCG-H repeatedly indicated quiescent GTD, suggesting that therapy should be halted. This was not done (Table 5).

Transformation from quiescent GTD then occurred, and for the first time, active GTN was apparent. This was demonstrated by a further referral and out finding of 28 mIU/ml hCG and for the first time of positive hCG-H results (15% of hCG). This was confirmed 1 month later by rising hCG (1312 and than 2710 mIU/ml) and by the detection of lung metastases. At this time, having assumed resistance to so many different combination chemotherapy regimens, it was decided to use a bleomycin-based protocol for treating the disease. The patient developed pulmonary fibrosis as a complication of the bleomycin administration and subsequently died.

Case 3, false-positive hCG

The patient had persistent low levels of hCG of approximately 120 mIU/ml lasting for 10 months. During this period, she received single agent methotrexate and actinomycin D. She then underwent a hysterectomy. Neither chemotherapy nor surgery had an impact on the hCG level. She was referred to the Service, and the absence of hCG immunoreactivity in parallel urine samples, the widely varying hCG results in different assays, and the suppression of immunoreactivity with a heterophilic antibody blocking agent together confirmed that she had false-positive hCG. All treatment of this patent was then stopped. We were informed that the false-positive hCG immunoreactivity disappeared over 6 months (Table 6).

Discussion

Here, we present an overview of 170 patients presenting with low levels of hCG, persisting for 3 months of longer.

Table 5
Complex case of quiescent GTD treated without success by numerous chemotherapy regimens and hysterectomy during 3 years with quiescent GTD

Month	hCG mIU/ml	Progress and comments
0		Incidental home pregnancy test. Ectopic pregnancy shown by ultrasound
0	278	Laparoscopic left salpingectomy. Fetal sac with necrotic fetal and placental tissue (yellowish mass) observed in pathology. On periphery, multinucleated cells observed without villous stroma. Ectopic choriocarcinoma suggested
0	130	MRI/CT of head, chest, and pelvis is unremarkable
		Formal diagnosis: non-metastatic choriocarcinoma
0	90	Methotrexate 14 regimens started
1	11	
6	8.5	At completion of 14th regimen methotrexate
8	19	Start on 5 EMA-CO (etoposide/methotrexate/actinomycin D, alternating weekly with cyclophosphamide and vincristine) regimens
11	12	Start on 5 MAC weekly regimens
12		CT reveals mass at original salpingectomy site, transabdominal hysterectomy, and salpingo-ooporectomy ordered. Giant multinucleated cells observed with large zones of tumor necrosis. Tumor is not hemorrhagic. Viable tissue limited to the core of the tumor. Ectopic choriocarcinoma again suggested
13	6	CT and PET scans of pelvis are unremarkable
15	6	Start on taxol–cisplatin 4-weekly regimens
15	7.7	Referral to hCG Ref. Service, quiescent GTD, halt therapy concluded but ignored
16	22	Start on 13 EMA-EP (etoposide/methotrexate/actinomycin D, alternating weekly with etoposide and cisplatin) regimens
25	14	After 13 EMA-EP regimens
26	4	Start on 3 VIP (oral etoposide/ifososamide/cisplatin) 3-day regimen
27	15	Start on 5 Xeloda monthly regimen programs
32	21	Completion of Xeloda, MRI/CT of head, chest, and pelvis are unremarkable
34	36	Start of 3 hydroxyurea regimens
36	28	Referral to hCG Ref. Service, quiescent GTD, halt therapy concluded but ignored
36	36	Start 4 VPB (vinblasine/cisplatin/bleomycin) regimens
37	1312	Referral to hCG Ref. Service, transformed quiescent GTD, active disease reported
37	1350	Start further 5 VPB regimens
38	2710	MRI of head and CT of chest reveal lung nodules. Development of pulmonary fibrosis as complication of bleomycin chemotherapy, patient expires

This led to active GTN and to patient's demise. Patient age 35, para 3, gravida 4, case started by incidental pregnancy test. All hCG data are from patients records, or testing institution, except as indicated.

t6.1 Table 6
t6.2 Representative case of false-positive hCG persisting for 10 months, treated without success by chemotherapy regimens and hysterectomy

t6.3 Month	hCG mIU/ml	Progress and comments
t6.4 0	116	Incidental pregnancy test prior to minor surgery
t6.5 0	126	Dilation and curettage reveal no evidence of pregnancy
t6.6 0	168	Ultrasound and laparoscopy reveal no ectopic pregnancy
t6.7 1	116	
t6.8 4	110	MRI/CT of head, chest, and pelvis is unremarkable
t6.9 5	134	
t6.10 5	120	Start 4 methotrexate regimens
t6.11 5	124	
t6.12 6	113	CT of pelvis shoes uterine irregularities, CT of chest unremarkable
t6.13 7	120	Start actinomycin D bolus regimen
t6.14 7	134	MRI of pelvis indicates uterine irregularities
t6.15 9	124	Hysterectomy, pathology is unremarkable
t6.16 10	142	
t6.17 10	0	Referral to hCG Ref. Service, false-positive hCG demonstrated, all therapy halted

t6.18 Patient age 34, para 2, gravida 2, case started by incidental pregnancy test. All hCG data are from patients records, or testing institution, accept as indicated.

Over half of these patients received chemotherapy and/or hysterectomy. The therapy was ineffective, demonstrating the futility of treatment in non-threatening diseases. Only 13 of 170 or 7.6% of patients had an actual malignancy. The vast majority (157 cases) had false-positive hCG results, quiescent GTD, or pituitary hCG. False-positive hCG occurs when human heterophilic antibodies in the circulation interfere with hCG tests, cross-linking antibodies [1–4]. Quiescent GTD is a benign form of trophoblast disease marked by the absence of cytotrophoblast cells, the invasive or malignant cell component [6,9]. The gonadotrope cells of the anterior pituitary are under the control of a hypothalamic releasing factor which is normally controlled, limiting gonadotropin release, by sex steroids during the menstrual cycle. When a woman is in perimenopause or post-menopause, the limited production of sex steroids causes maximum gonadotropin release. This can be accompanied by the production of some hCG by the gonadotrope cells. As such, low level hCG production is normal in these older women.

How commonly do patients present with persistent low hCG? In the Service experience, approximately one third of cases occur in patients with GTD/GTN history (n = 59) and approximately two thirds in those with no history (n = 111). The University of New Mexico Department of Obstetrics and Gynecology refers all cases of persistent low hCG to the Service. Based on the Service's experience and that at Yale (both of which are limited but institutionally comprehensive), we estimate that approximately one in eight GTD cases at some point develops persistent low levels of hCG (Cole LA and Kohorn EI, unpublished data). Based on the incidence of hydatidiform mole in the USA [8], this would broadly suggests approximately 500–1000 cases of persistent low hCG in patients with history of GTD in the USA. This is the best estimate available at this time. If this represents one third of the persistent low hCG patients, then there are possibly 1500–3000 total cases in the USA each year.

Clinical practice dictates treatment of patients with elevated hCG in the absence of pregnancy for GTN or recurrent GTN. Clinicians generally believe that the presence of any "true" hCG indicates that the patient has choriocarcinoma or GTN, even if there is no clinical evidence for this. As shown here and by others, in the vast majority cases with persistent low hCG, all therapies are ineffective [2–5,11,12]. Data presented here show that only 7.6% have a malignancy and need chemotherapy, yet over half of these patients received the chemotherapy or surgery for assumed GTN or non-trophoblastic malignancy (89 of 170). Better understanding of this spectrum of diseases and the limitations of laboratory testing is critical in preventing unnecessary treatments that can be fatal.

Three case studies that illustrate the consequences of over treatment of persistent low hCG have been presented. In all 3 cases, healthy women had hysterectomies and chemotherapy, which were ineffective. One woman died after numerous chemotherapy regimens were used unsuccessfully to treat quiescent GTD. When she later developed active GTN, other therapies were considered. She died as a complication of seldom used bleomycin.

Informed care is needed in the management of patients with persistent low levels of hCG.

Based on the Service's experience, the following guidelines are proposed for the management of all patients with persistent low level hCG, including those following hydatidiform mole or GTN/choriocarcinoma.

1. After ruling out pregnancy and ectopic pregnancy, and in all patients with a history of GTD or GTN, start by determining if the hCG is biologically real. False-positive hCG is a common cause of persistent low levels of hCG. The limitations of the common laboratory assays need to be understood. These are the Abbott AxSym, Bayer ASC180, Bayer Centaur, Ortho Vitros, and Beckman Access serum hCG tests (5 of the most commonly used tests in the USA). These commercial tests have a proven propensity to give false-positive hCG results. All competitive hCG assays or radioimmunoassays have an inherent problem with false-positive hCG results [1,10,13–16]. Particular care is needed when these assays are used. In all cases, the serum should be sent to an alternative laboratory, one using the Abbott Architect, Roche Elecsys, or DPC Immulite, tests that have not been reported to give false-positive results. If the alternative test shows a similar hCG result, then it is a valid hCG result.
2. If the hCG is real hCG, it is important to determine if active GTN, PSTT, or non-trophoblastic malignancy is present. In our experience, this is unlikely since it represents a very small proportion or approximately 7.6% of cases, but it must be checked. The serum samples should be to sent to specialist laboratories (for instance, Nichols Reference Laboratories of Quest Diagnostics or the USA hCG Reference Service Reference Service) for measurement of hCG-H (if hCG-H is detectable, >1 ng/ml, then active GTN/

choriocarcinoma may be present [6]) and hCG free β-subunit (hCG free β-subunit results are in ng/ml, multiply by 17 to convert to molar equivalent of hCG in mIU/ml [7]). If hCG free β-subunit is more that one third of hCG result, then PSTT or non-trophoblastic malignancy is likely [7]. If neither hCG-H nor significant hCG free β-subunit is present, then this is very likely a case of quiescent GTD. If the patient is peri- or post-menopause, or has had an oophorectomy, then pituitary hCG is likely. In that case, the patient should take hormone replacement therapy or oral contraceptives. After 2–3 weeks, this should suppress hCG production if it is of pituitary origin.

These guidelines will help to differentiate non-malignant states from a malignancy using readily available biochemical tests before starting any therapy. This is equally applicable whether a patient is identified by an incidental pregnancy test or is actively being monitored for gestational trophoblastic disease (Table 1). The USA hCG Reference Service experience has 170 cases with persistent low levels of hCG persisting for 3 months or longer, with no evidence of pregnancy or imaging evidence of tumor. The 71 cases diagnosed as having false-positive hCG were based on multiple observations by the USA hCG Reference Service. These are: 1. the presence of hCG immunoreactivity in serum but not urine; 2. varying hCG results (more than 5-fold) or negative results in 3 or more hCG tests; 3. the suppression of result by a heterophilic antibody blocking agent. The 17 pituitary hCG cases were defined as those peri- or post-menopausal women with low levels of hCG <25 mIU/ml, having excluded false-positive hCG, non-trophoblastic malignancy and choriocarcinoma/GTN or placental site trophoblastic malignancy. The 69 cases with quiescent GTD and the 11 cases with PSTT are those described in preceding publications [6,7]. All hCG values are those determined at the USA hCG Reference Service.

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