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Evaluation of hPG80 (Circulating Progastrin) As a Multi-Cancer Early Detection (MCED) Test In a Private Clinic in Norway

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Abstract

Background: Early cancer detection is crucial for effective treatment. In a private clinic in Norway, we evaluated hPG80 (circulating progastrin), a novel blood-based biomarker, as a potential multi-cancer early detection (MCED) test in individuals over 50 at risk for cancer.

Methods: Plasma EDTA samples from 24 asymptomatic patients were analyzed using the DxPG80.lab ELISA kit (Biodena Care, France) between January and March 2020. Participants were stratified based on hPG80 levels: low (below Limit of Quantification), intermediate (above LoQ but below Upper Limit of Normal), and high (above ULN).

Results: Three early-stage cancers (colorectal, prostate, and lung) were detected in individuals with intermediate or high hPG80 levels. All were treated successfully, with no relapses after four years. One patient with high hPG80 levels was initially diagnosed with a premalignant kidney cyst and later developed two separate lung cancers, which were also successfully treated. Four other patients with elevated hPG80 underwent further screening, but no malignancy was found; they remain cancer-free after follow-up. Sixteen patients with low hPG80 levels have not developed cancer over four years. The study showed a sensitivity of 100%, specificity of 80%, Negative Predictive Value (NPV) of 100%, and Positive Predictive Value (PPV) of 50%.

Conclusion: hPG80 may detect cancer earlier than conventional methods, supporting its role as a promising MCED tool. Low levels were associated with no cancer development, suggesting hPG80's potential use for risk stratification and tailored monitoring in clinical settings.

Keywords: hPG80; MCED Test; Blood-based biomarker.

Introduction

One of the most promising trends in improving survival of cancer is the emerging use of tests designed to detect cancers early, Multi-Cancer Early Detection Tests (MCED). By prioritizing earlier action, early detection can shift the average diagnosis from advanced to localized stages. For patients over 40, implementing these testing in the US could prevent an estimated 66,000 cancer deaths annually [1]. There are several MCED tests available on the market and a huge number in the pipelines [2].

Recently, the blood-based biomarker, hPG80 (circulating progastrin), has been shown to be a promising diagnostic biomarker for various types of solid tumours [3-9]. In physiological conditions, progastrin is the precursor of the gastrointestinal hormone, gastrin, synthetized by antrum G cells and maturated into gastrin during digestion10. In pathological conditions, the GAST gene, which encodes progastrin, is a direct target of oncogenic pathways frequently activated in various types of cancers such as the APC/β-catenin or Ras pathways [11,12]. In these cancer cells, progastrin is not maturated into gastrin and is released as such. Once released into the blood stream, circulating progastrin is named hPG80 to distinguish it from the precursor of gastrin. Many studies have demonstrated that hPG80 plays important roles in various pathological processes including cell proliferation, disruption of cell junctions, inhibition of apoptosis, survival of cancer stem cells and angiogenesis [13-22]. High hPG80 concentrations have been reported in various types of cancers [3-9]. Additionally, higher hPG80 levels were closely associated with a worse prognosis in metastatic renal cell carcinoma, hepatocellular carcinoma, breast cancer and glioblastoma [4,5,7,8].

The risk of developing malignant disease rises significantly after the age of 50. To explore the potential of hPG80 in detecting early, undiagnosed cancers, we evaluated hPG80 in patients over 50 years of age during a three-month inclusion period in early 2020. The primary inclusion criterion was the absence of symptoms or clinical signs suggesting a possible cancer diagnosis.

Materials and Methods

hPG80 level measurements in the blood samples

The ELISA DxPG80.lab kit (Biodena Care, Grabels, France) was used to measure hPG80 levels in plasma samples according to the manufacturer's instructions described previously [23]. The analytical performances of the kit are described in Cappellini et al. [23]. Briefly, the limit of quantitation (LoQ) was 3.3 pM and the upper limit of normal (ULN) was 10.9 pM. The inter- and intra-assay coefficients of variation (CV%) were <10%. No cross-reactivity was detected with gastrin-17, gastrin-Gly, or C-terminus flanking peptide. No cross-reactivity was detected with other blood biomarkers, such as cancer antigen 125, carcinoembryonic antigen, or prostate-specific antigen. No interference was detected with chemicals, such as SN-38 and 5 fluorouracil, or with triglycerides, cholesterol, or hemoglobin.

Individuals enrolled in this real-world study

Our private medical clinic provides services in general medicine, internal medicine, and occupational medicine, catering to self-paying patients. During a three-month period in early 2020, we tested hPG80 in patients with the following inclusion criteria: age above 50 and no suspicion of malignancy (i.e., absence of symptoms or clinical signs of cancer). All patients were examined clinically and signed an informed consent agreement.

We ranked the individuals according to risk. Normal risk was defined as age only for both sexes, age above 50. The following qualified for high risk: a) a first-degree relative with proven cancer b) a previous history of cancer or c) a smoking history of more than 20 years.

Individuals were stratified as low levels of hPG80 when levels were below the LoQ (3.3 pM), intermediate levels of hPG80 when levels were above the LoQ and below the ULN (10.9 pM), and high levels of hPG80 when levels were above the ULN.

Individuals with high levels of hPG80 were further examined (clinical examination, laboratory testing and Computed Tomography (CT) scans) for presence of an unknown cancer. A standard clinical examination was extended to include breast palpation for females, a full skin examination looking for melanomas of both sexes, scrotum palpation for males and rectal exploration for both sexes. Laboratory examination included regular hematology, liver and kidney tests, lactate dehydrogenase as well as the tumour markers PSA, CEA, CA 19.9, CA 125 (females) and PSA (males). The values of the tumour markers are mentioned in the case report where relevant. Patients were also tested for blood in urine and stool. CT scan with contrast included neck, chest, abdominal and pelvic area. In the case of no finding, patients were referred to colonoscopy. For those where a cancer was detected, they were treated and followed further by the hospital. In case of no findings, we performed a clinical and laboratory evaluation every year. They were informed that every cancer symptom should lead to immediate action. For the intermediate level hPG80 group, we performed the same clinical examination and laboratory testing. If negative they were invited for a new clinical and laboratory examination after one year, including a new hPG80 testing. They were also informed about the importance of a rapid response if developing cancer symptoms. For the low level hPG80 group, a full clinical examination was performed. They were also informed to act fast if any cancer symptoms developed.

Initially it was planned to perform a structured follow-up with longitudinal analyses of hPG80. Due to the pandemics, there were strong restrictions of population movements in Norway, and most clinics were closed to non-acute medical services for several and long periods of time. Our plan of these longitudinal analyses had to be cancelled as well as the inclusion of further patients.

The follow-up of the population was performed late 2024/early 2025 after about 4 years. For those patients still visiting our clinic, clinical and laboratory investigations were performed. For those patients that had moved or visiting other clinics, their cancer situation was updated by calling them for an interview and having accept to consult their new physicians.

Statistical analysis

Data is expressed as median ± interquartile range (IQR) and mean ± standard error of the mean (SE). Differences in hPG80 levels were evaluated using the non-parametric Mann-Whitney U test. The diagnostic discriminative accuracy of hPG80 levels in patients with cancer compared to healthy subjects was assessed using Receiver Operating Characteristics (ROC) curve analyzes. Prism software (GraphPad Prism version 9.4) was used to perform all the statistical analysis and to create figures. The level of significance was set at $P<0.05$.

Results

Clinical characteristics

The population included 24 consecutive individuals without cancer suspicion (Table 1). The population included 5/24 (20.8%) females and 19/24 (79.2%) males with a median age of 58.5 yo (IQR 53-68 yo; range between 49 and 82 yo). 16/24 individuals (66.6%) had low levels of hPG80, 4/24 (16.7%) had intermediate levels and 4/24 (16.7%) had high levels. 9/24 of them (37.5%) were classified as high-risk patients and 15/24 (62.5%) as normal risk.

Patient	Gender	Age	Risk level	hPG80 (pM) at inclusion	Outcome	Category
Patient 1	Male	71	High risk	21	Complex Bosniac cyst and two separate lung cancers	High or intermediate levels of hPG80. Cancers detected.
Patient 2	Male	60	High risk	11.9	Lung cancer and benign colon polyps	
Patient 3	Female	65	High risk	9.4	Colon cancer and adenomas in colon	
Patient 4	Male	63	High risk	3.5	Prostate cancer	
Patient 5	Male	72	Normal risk	41	All examinations negative	High or intermediate levels of hPG80. No cancers detected after 4 years
Patient 6	Male	55	Normal risk	19.8	All examinations negative	
Patient 7	Male	49	Normal risk	5.8	All examinations negative	
Patient 8	Female	60	Normal risk	3.9	All examinations negative	

Patient 9	Male	56	Normal risk	<LoQ	No examinations	Negative hPG80. No cancer detected after 4 years
Patient 10	Female	82	Normal risk	<LoQ	No examinations	
Patient 11	Male	53	Normal risk	<LoQ	No examinations	
Patient 12	Male	69	Normal risk	<LoQ	No examinations	
Patient 13	Male	54	Normal risk	<LoQ	No examinations	
Patient 14	Male	51	Normal risk	<LoQ	No examinations	
Patient 15	Female	64	High risk	<LoQ	No examinations	
Patient 16	Male	75	High risk	<LoQ	No examinations	
Patient 17	Male	54	High risk	<LoQ	No examinations	
Patient 18	Male	53	Normal risk	<LoQ	No examinations	
Patient 19	Female	52	Normal risk	<LoQ	No examinations	
Patient 20	Male	57	Normal risk	<LoQ	No examinations	
Patient 21	Male	52	Normal risk	<LoQ	No examinations	
Patient 22	Male	53	High risk	<LoQ	No examinations	
Patient 23	Male 75	75	Normal risk	<LoQ	No examinations	
Patient 24	Male 62	62	High risk	<LoQ	No examinations	

Table 1: Summary of the clinical information from the 24 individuals included in the study.

4/24 patients (16.7%) were diagnosed with cancer within the 4-year follow-up period (3 were diagnosed shortly after testing). All 4 cancer cases were in the high-risk group. 4/24 individuals (16.7%) with high or intermediate hPG80 levels did not develop cancer during the 4-year follow-up. 16/16 individuals (100%) with low hPG80 levels remained cancer-free, including 11/16 (68.8%) in the normal-risk group and 5/16 (31.2%) in the high-risk group.

hPG80 Levels in cancer-free and cancer positive individuals and diagnostic performance

Plasma hPG80 levels in cancer-free and cancer positive individuals are shown in Figure 1 and Table 2. hPG80 levels were found to be significantly higher in cancer patients than in cancer free individuals (median: 10.65 pM, IQR: 3.50-18.73 vs 2.30 pM, IQR: 1.60-2.70, $P<0.0128$, respectively). Next, we conducted ROC curve analysis to assess the performance of hPG80 for differentiating between cancer positive and cancer-free individuals. As shown in Figure 2, the AUC value was 0.89 (95% CI = 0.75-1.00). Using a cut-off value based on the limit of quantification (LoQ) of the kit (3.3 pM), we found a sensitivity of 100%, a specificity of 80%, a negative predictive value (NPV) of 100% and positive prediction value (PPV) of 50%, to differentiate cancer positive from cancer-free individuals.

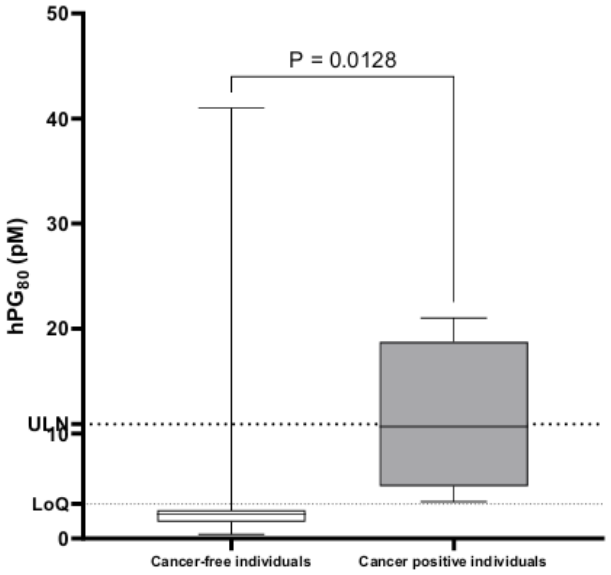

Figure 1: Plasma hPG80 levels in cancer-free and cancer positives individuals.

	Cancer-free individuals	Cancer positive individuals
Number of values	20	4
Minimum	0.4	3.5
25% Percentile	1.6	4.98
Median	2.3	10.65
75% Percentile	2.7	18.73
Maximum	41	21
Range	40.6	17.5
Mean	5.06	11.45
Std. Deviation	9.39	7.28
Std. Error of Mean	2.1	3.64

Table 2: Plasma hPG80 levels in cancer-free and cancer positive individuals.

Box-whisker plots show hPG80 levels in cancer-free individuals (n=20), in cancer positives individuals (n=4). Boxes represent the interquartile range, and the horizontal line across each box indicates median values. The statistical differences were evaluated with the Mann Whitney U test. LoQ: limit of Quantification and ULN: Upper Limit of Normal.

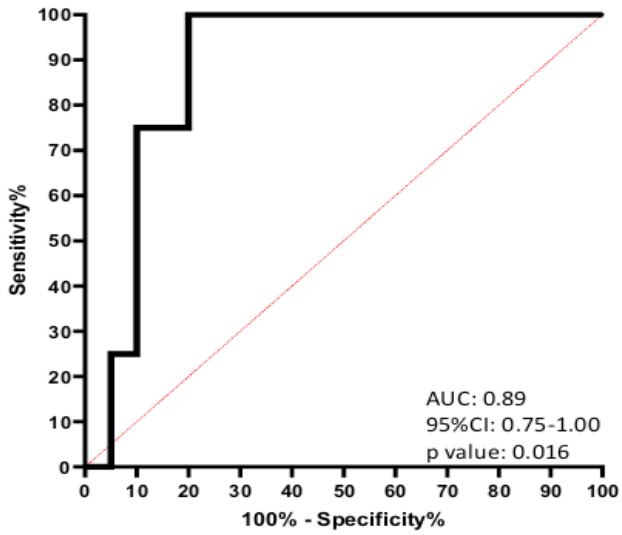

Figure 2: Diagnostic performance of hPG80 for cancer positive individuals. Receiver operating characteristic curves (ROC) of hPG80 in differentiating cancer positive individuals from cancer-free individuals. AUC: Area under the curve and 95% CI: 95% confidence interval.

Cases Presentation

Patient 1.

A 71-year-old male, healthy and fit, with a 50-year smoking history, consulted our clinic in February 2020. His family history included gastric cancer in his father (deceased at 73) and four siblings diagnosed with cancer: two with leukemia, one with prostate cancer, and one with rectal cancer - all successfully treated and alive at the time being. Based on our risk stratification criteria, he was classified as high-risk for cancer development. His hPG80 level was 21 pM, prompting a comprehensive clinical and laboratory assessment, including a CT scan of the thorax and abdomen. All laboratory results were normal (including PSA 3.9 ng/mL and CEA of 2.1 ng/mL), and his scan of thorax was normal. However, a complex cyst (29 × 21 × 22 mm) with a thick, irregular wall and contrast uptake was detected in his left kidney, classified as a Bosniak type 3 cyst. He was referred to a urology hospital for follow-up imaging, as per guidelines. After 2 years, his cyst was surgically removed.

In April 2024, he developed a persistent cough, leading to a new CT scan, which revealed two separate lung cancers: a squamous cell carcinoma in the right middle lobe (treated with surgery) and an adenocarcinoma in the left upper lobe (treated with curative-intent stereotactic radiation). He subsequently underwent adjuvant chemotherapy and immunotherapy for both tumors. As of now (11 months post-diagnosis), he remains disease-free and has returned to work.

Patient 2.

A 60-year-old male with a medical history of percutaneous coronary intervention (PCI) of the LAD and Cx in 2018, type 2 diabetes, and hypothyroidism (managed with oral antidiabetic drugs and thyroxin) consulted our clinic in February 2020. He was classified as high-risk for cancer due to a long smoking history, although he had stopped smoking 16 years prior and had no first-degree relatives with cancer. His hPG80 level was 11.9 pM, prompting a comprehensive evaluation, including: full clinical and laboratory assessment (all normal and PSA 2.16 ng/mL and CEA of 3.4 ng/mL), CT scan of the thorax and abdomen and colonoscopy. CT scan showed a small nodule in the right lower lobe, and an 11 mm nodule in the left lower lobe, both initially considered benign post-infectious nodes. The radiologist recommended a follow-up CT in 3 months. The colonoscopy detected seven benign polyps which were subsequently removed. Given his elevated hPG80 level, we referred him to a PET scan, which showed moderate uptake in the left lower lobe nodule. A biopsy was inconclusive, leading to thoracotomy in July 2020. Intraoperative biopsy confirmed a carcinoid tumor, prompting an onsite lobectomy of the left lower lobe. Final histology confirmed a completely resected carcinoid tumor with no lymph node metastases. The patient recovered well and was in good condition 10 weeks post-surgery. Four years later, follow-up at the hospital has shown no recurrence of lung cancer and no signs of other malignancies.

Patient 3.

A 65-year-old female, healthy and never-smoker, consulted our clinic in February 2020. Her mother had been diagnosed with colon cancer at age 78, classifying her as high-risk due to a first-degree relative with cancer. Her hPG80 level was 9.4 pM, prompting further evaluation. Full clinical examination and laboratory tests revealed low hemoglobin levels, positive fecal occult blood (FOB) test, and negative tumour markers including CEA of 1.8 ng/mL. Colonoscopy detected an obstructive tumor in the transverse colon and an adenoma at the anorectal junction. She underwent successful surgery, her tumour was classified as Duke C, followed by adjuvant chemotherapy (FLOX) for 3 months. Follow-up results showed that her hPG80 level dropped below the LoQ. No signs of recurrence or new cancers have been detected after 4 years of observation.

Patient 4.

A 63-year-old male, healthy never-smoker, consulted our clinic in February 2020. His only medical issue was enlarging prostate symptoms for several years, with a normal PSA of 1.2 ng/mL. His family history included one sister, and one daughter diagnosed

with breast cancer (both successfully treated with surgery, adjuvant radio/chemotherapy, and no signs of recurrence). Given his two first-degree relatives diagnosed with cancer at a young age, he was categorized as high-risk for cancer development. Initial findings showed an hPG80 level of 3.5 pM (intermediate range). He was advised to repeat hPG80 testing after 12 months. In August 2020, the patient underwent transurethral resection of the prostate (TUR-P) due to worsening lower urinary tract symptoms (LUTS). Preoperative ultrasound showed a normal prostate. PSA level was stable at 1.2 ng/mL. The biopsy showed however an unexpected result: a prostate cancer (Gleason 3+3 = 6). TUR-P was successful, resolving urinary symptoms. Three months post-treatment, PSA decreased to 0.66 ng/mL, hPG80 level fell below the LoQ, indicating biochemical success. Four-year follow-up showed no signs of cancer recurrence. PSA was stable at 0.7 ng/ml and hPG80 remained below LoQ.

Discussion

In this first monocentric real-world data study with a 4-year follow-up, we were able to detect 4 patients with cancers of different origins using hPG80 test that would not have been detected that early using standard procedures. Furthermore, the 16 patients with levels of hPG80 below LoQ were cancer-free 4 years after the initial enrolment. Overall, this observational study yielded a sensitivity of 100%, a specificity of 80%, a Negative Predictive Value (NPV) of 100% and a Positive Predictive Value (PPV) of 50%. Of course, this study has limitations. First the small number of patients enrolled in the study can limit the power of the statistical analysis. Second, this study is a single-centre study that will require external validation to confirm these promising results.

New cancer screening technologies, like the MCED tests, should be carefully evaluated for efficacy and cost, but adding it to current guidelines could be an efficient strategy for high-risk groups (e.g., older adults, smokers) to improve detection of multiple cancer types, including aggressive ones without current screening options [1,2,24-26].

Who should be screened with hPG80 and how often?

Since there are no established studies determining the optimal screening population or interval for hPG80 or similar MCED tests, we can use cancer registry data to refine our recommendations until larger screening studies are conducted. Norway's Cancer Registry has maintained high quality registration and follow-up of all newly diagnosed cancers for many years. In 2023, 38,094 new cancer cases were recorded, with 17,708 cases (46.4%) in women and 20,386 cases (53.6%) in men [27].

Between 2018 and 2023, the age distribution of cancer incidence revealed that only 7% of all cancers in men occur before age 50. In women, 13.3% of cancers are diagnosed before age 50. After age 50, there is a sharp rise in cancer incidence, particularly in men. In men, 72.9% of all cancers occur between ages 50 and 80. In women, 64.5% of all cancers occur within this age range. Additionally, there has been a notable rise in early-onset cancers, a trend expected to continue in the coming years [27].

Screening recommendations should be based on risk group. The normal-risk group criteria include no prior malignancy, no significant smoking history, no first-degree relatives with cancer diagnosed at a young age. Screening recommendation for this group is to start screening at age 50 and continue until age 80. The hPG80 test should be done every two years (biennial screening). The high-risk group criteria include history of previous malignancy, smoking history (current or former heavy smoker), and first-degree relatives with cancer, particularly if diagnosed at a young age. Screening recommendation for this group is to start screening at age 40. The hPG80 test should be done once a year between age 40 and 50, and further testing after 50 every other year. These recommendations are based on cancer registry data and current understanding of the biology of cancer progression. Future large-scale screening studies will help further refine these guidelines.

hPG80-based screening and follow-up protocols

hPG80 values are classified as follows: elevated above the ULN, intermediate between the LoQ and ULN, low below the LoQ. As clinicians, the use of MCED tests and subsequent follow-up must be assessed from both a clinical benefit and economic feasibility standpoint. Based on years of clinical experience and findings from this observational study, we propose the following screening and follow-up protocols (Table 3).

Reference Range	Levels	Result	Protocol & Action
Up to LoQ	low	Not Quantifiable	No Action / Repeat Annual Prefered
LoQ – ULN	intermediate	Quantifiable and Significant "Grey Zone"	Blood based biomarker dosage – if nothing repeat the test in 12 months – Under Surveilliance
> ULN	high	Quantifiable and High	Localization Protocol

Table 3: potential algorithm using hPG80.

hPG80 level above ULN (elevated levels - high risk):

We recommend a comprehensive clinical examination focusing on cancer, along with blood tests to detect cancer markers and checks for blood in urine and feces. Additionally, a CT scan of the neck, chest, abdomen and pelvis should be performed. If no abnormalities are found, we suggest a colonoscopy for further evaluation. If any findings are detected, the next steps will depend on the nature of the findings. If no issues are identified, the patient should be clearly informed of their high-risk status for developing cancer and advised to promptly seek medical attention if any cancer-related symptoms arise. We recommend annual follow-up with clinical and laboratory evaluations. At this stage, additional hPG80 testing is not necessary since the patient has already been classified as high-risk. In the event of a cancer diagnosis, hPG80 should be tested for 3 months post-treatment to confirm the biochemical success of the treatment.

- hPG80 between LoQ and ULN (intermediate levels – moderate risk):

We recommend a comprehensive clinical examination with focus on cancer detection: blood tests including tumour markers, urine and faecal blood tests. If abnormalities are found, further evaluation is conducted accordingly. If no malignancy is detected, repeat hPG80 testing in 12 months. Further follow-up depends on hPG80 progression and patient history. We recommend annual clinical and laboratory examinations with a focus on early malignancy detection.

- hPG80 below LoQ (low levels – low risk):

A routine clinical examination at the scheduled screening testing is recommended. If no abnormalities are found, no additional testing is required. A new hPG80 test should be repeated every 2 years after age of 50. Since hPG80 does not detect all cancers, patients should be informed of cancer warning signs and advised to seek medical attention if symptoms arise.

These recommendations provide a structured, evidence-based approach to early cancer detection while ensuring efficient use of medical resources.

In our small observational study, the mean age at testing was 58.5 years. According to data from the Norwegian Cancer Registry, the annual risk of a 58-year-old being diagnosed with cancer is 0.075%. However, in our study population, we identified three cancers at the time of hPG80 testing, meaning 12% of our participants were diagnosed with cancer. While these numbers are based on a small sample size and are not statistically significant, we were struck by the unexpectedly high number of cancers detected. Furthermore, our initial plan was to conduct follow-up hPG80 testing for all patients with intermediate levels after 12 months. Unfortunately, this had to be canceled due to the COVID-19 lockdown.

Conclusion

Beyond its well-established role as a potential tumor marker with clinical benefits in cancer monitoring, prognosis, and early relapse detection, we now use hPG80 as an MCED test following the guidelines outlined above. Through this experience, we have learned the importance of closely following patients with high or intermediate hPG80 levels. Integrating hPG80 testing into our existing check-up protocol has significantly enhanced early cancer detection, reinforcing its value as a promising tool in preventive healthcare.

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Ethical Consideration

Informed consents were obtained from all subjects.

Conflict of Interests

AP is Chief Scientific Officer of Progastrin Manufacturing.

DJ is senior scientist consultant of Progastrin Manufacturing.

All other authors: No conflict of interest.

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