Basal Plasma Levels of Copeptin are Elevated in Inactive Inflammatory Bowel Disease after Bowel Resection.

Ohlsson, Bodil; Melander, Olle

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Basal Plasma Levels of Copeptin are Elevated in Inactive Inflammatory Bowel Disease after Bowel Resection

Bodil Ohlsson and Olle Melander

Department of Clinical Sciences, Section of Internal Medicine, Skåne University Hospital, Malmö, and Lund University, Lund, Sweden.

ABSTRACT: Evidence of interactions between the enteric nervous system, neuropeptides, and the immune system is growing. The aim of this study was to examine basal plasma levels of a variety of peptide precursors in patients with inflammatory bowel disease (IBD). In two middle-aged cohorts, Malmö Preventive Medicine (n = 5,415) and Malmö Diet and Cost Study (n = 6,103), individuals with the diagnosis of IBD were identified. Medical records were scrutinized. Three controls were matched for each patient. Copeptin, midregional fragments of adrenomedullin, pro-atrial natriuretic peptide, and proenkephalin A, as well as N-terminal protachykinin A and proneurotensin were analyzed in the plasma. Sixty-two IBD patients were identified. The only difference between patients and controls was higher copeptin levels in the patients compared with controls (P = 0.006), with higher copeptin levels in resected than unresected patients (P = 0.020). There was no difference in any precursor levels between Crohn's disease and ulcerative colitis, between different distributions of disease lesions, or between different treatments.

KEYWORDS: copeptin, inflammatory bowel disease, irritable bowel syndrome-like symptoms, neuropeptides, precursors

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CORRESPONDENCE: bodil.ohlsson@med.lu.se

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Introduction

Interactions between the enteric nervous system (ENS) and the immune system have been the subject of much new evidence in recent years. Inflammatory cells in the intestine express receptors for neuropeptides, and the enteric neurons are responsive to cytokines secreted from inflammatory cells.[1,2] Nevertheless, the focus in inflammatory bowel disease (IBD) has been the immune system, and few studies have examined the role of neuropeptides in the development and maintenance of the disease, especially in inactive disease. Even when the disease is under control and no signs of inflammation are present, patients with IBD often suffer from abdominal pain and functional abdominal complaints, so-called irritable bowel syndrome (IBS)-like symptoms.[3] The reason for these complaints is unknown, but hypersensitivity, low-grade inflammation, or impaired epithelial barrier has been suggested.[3] Imbalance in neurotransmitters/neuropeptides has only been sparsely examined in relation to hypersensitivity and IBS-like symptoms. Structural changes occur in the ENS during IBD,[4] and the precursor mRNAs encoding substance P and related receptors have been found to be upregulated in Crohn's disease.[5,6] Recently, lower plasma levels of enkephalins were described in active IBD patients compared with controls.[7]

Adrenomedullin, enkephalin A, neurotensin, and substance P, as well as their receptors, have been demonstrated in the gastrointestinal tract[2,8–11] and found to be involved in inflammation and nociception.[10–14] Adrenomedullin has been shown to ameliorate the induction of colitis in animal models and to induce wound healing.[10] Enkephalins exert both anti-inflammatory and anti-nociceptive effects in induced colitis in animal models.[13] Endogenous neurotensin facilitates visceral pain responses and is necessary for the development of irritant-induced hyperalgesia.[12] Substance P is involved in the regulation of mast cell activation,[14] and the upregulation of substance P observed in the gastrointestinal tract in IBD patients seems to be associated with ongoing local inflammation, which can be clinically quiet.[11] Copeptin, the precursor of arginine vasopressin (AVP), is secreted from the posterior pituitary gland, and all three vasopressin receptors have been found in the gastrointestinal tract.[15] The main roles of vasopressin are water reabsorption through V2 receptors,[16] regulation of vascular tone by V1a receptor activation,[17] and physiological adaptations to stress through adrenocorticotropic hormone (ACTH) release by the posterior pituitary involving V1b receptors.[18] A few studies have examined the pharmacological, but not the physiological, effect of vasopressin on water and sodium absorption and secretion in human intestines.[19,20] In rodents, vasopressin has been reported to exert proinflammatory effects through mast cell activation and enhanced epithelial permeability in the colon.[21] Atrial natriuretic peptide (ANP) is secreted from the heart and contributes to substantial fluid loss via the gastrointestinal tract.[22] Peptides have a short half-time in plasma, so it is advantageous to measure the highly stable precursors instead

of peptides, which are synthesized in stoichiometric amounts relative to the mature peptides.[23,24]

The aim of the present pilot study was to analyze the basal plasma levels of a variety of peptide precursors involved in the physiology of the gastrointestinal tract, ie, copeptin, midregional fragment of pro-adrenomedullin (MR-proADM), midregional fragment of pro-atrial natriuretic peptide (MR-proANP), midregional fragment of proenkephalin A (MR-PENK A), N-terminal protachykinin A (NT-PTA), and proneurotensin, in two well-defined populations of patients with IBD, as these precursors have been discussed in the development and activity of gastrointestinal diseases.

Material and Methods

The research protocol was in accordance with the Declaration of Helsinki, and the study was approved by the Ethics Committee of Lund University (2013/609).

Study population. The Malmö Preventive Medicine (MPM) study is a population-based, prospective, epidemiologic cohort of 22,444 men (born between 1921 and 1949) and 10,902 women (born between 1926 and 1949) from Malmö, Sweden, who underwent baseline examinations between 1974 and 1992.[25] From this cohort, 18,240 were re-examined from 2002 to 2006, and new blood samples were collected. Out of these, 5,415 were randomly chosen for precursor analyses.

The Malmö Diet and Cancer Study (MDCS) is a population-based, prospective, epidemiologic cohort of 28,449 men (born between 1923 and 1945) and women (born between 1923 and 1950) from Malmö, Sweden, who underwent baseline examinations between 1991 and 1996. From this cohort, 6,103 individuals were randomly selected to participate in the MDCS cardiovascular cohort (MDC-CC), which was designed to investigate the epidemiology of carotid artery disease between 1991 and 1994.[26] The baseline examination procedure has been described in detail previously.[27]

Briefly, the MPM and MDC-CC baseline examinations included a dietary assessment; a self-administered questionnaire about marital status, education, employment, smoking habits, wine consumption, physical activity, medical conditions and medication; anthropometric measurements; and collection of blood samples.

At baseline, no questions concerning bowel diseases were asked. Thus, we used local registers to retrieve prevalent cases of IBD according to the International Classification of Diseases (ICD) version 8, 9, and 10. We initially identified 35 patients with IBD in the MPM re-examination cohort with available precursors analyses, and 70 patients with IBD in the MDC-CC cohort. The diagnoses were validated by scrutinizing all the medical records, and all patients with a clinical diagnosis made after careful clinical examination including endoscopy and histopathological examination were included in the study. From the medical records, the activity and extent of disease, concomitant diseases, drug treatment, and bowel resections were recognized. Disease involvement in more than one gastrointestinal segment was defined as extensive disease. Six patients in the MPM cohort and nine patients in the MDC-CC were excluded due to wrong diagnosis, the coexistence of cardiovascular disease, diabetes mellitus, or malignancy, or the medical records not being found in the archive and hence validation according to prevalent or incident IBD, disease activity, and concomitant diseases could not be performed. Thus, after validation of the information from the register using the medical records, 22 prevalent and 7 incident cases of IBD remained in the MPM cohort and 40 prevalent and 21 incident cases in the MDC-CC, at the time point when plasma samples were collected.

Analyses. At the MPM reexamination and at the MDC-CC baseline examination, blood was collected in the morning after 12 hours of fasting, and plasma was separated and immediately frozen at −80°C. In the MDC-CC, plasma samples were available for analyses of MR-PENK, NT-PTA, and proneurotensin in 4,632 participants, and for analyses of copeptin, MR-proADM, and MR-proANP in 4,742 participants. The excluded participants (due to lack of plasma samples) were slightly younger, but did not otherwise differ in terms of sex, smoking habits, diabetes, hypertension status, body mass index (BMI), or plasma lipids.[27,28]

Copeptin, the stable precursor peptide of AVP, was measured by a murine monoclonal antibody directed to amino acids 137–144 of proAVP using a commercially available assay in the chemiluminescence/coated tube format (LUMItest CT-proAVP, Brahms GmbH) as described previously.[29]

MR-proADM and MR-proANP were analyzed using sandwich immunoluminometric assays targeted against amino acids in the mid-regions of the respective peptide (Brahms Sevadil LIA® and Brahms Seristra®, respectively, Brahms GmbH).[30,31] Briefly, two polyclonal antibodies targeted to amino acids 45–92 of proADM was used for the measurement of MR-proADM,[30] and polyclonal sheep antibodies specific for amino acids 73–90 were used for the measurement of MR-proANP.[31]

MR-PENK A is the stable fragment of the peptide precursor of enkephalins, and NT-PTA is the stable fragment of the peptide precursor of substance P. MR-PENK A was measured by a sensitive chemiluminescence immunoassay against amino acids 119–159 of the MR-PENK A precursor fragment (Brahms GmbH).[23] A similar sensitive chemiluminescence immunoassay was developed to detect amino acids 1–37 of NT-PTA (Brahms GmbH).[32]

Proneurotensin was measured by a recent developed chemiluminometric sandwich immunoassay to detect a proneurotensin precursor fragment (pro NT/NMN 1–117) (Brahms GmbH).[33]

Statistical analyses. Patients with valid, prevalent IBD in the MPM (n = 22) and MDC-CC (n = 40) cohorts were matched with three controls from their own cohort. Those subjects with prevalent cardiovascular disease, diabetes mellitus, or malignancy prior to the blood sampling were excluded from both patients and controls, as these diseases

may interact with plasma precursor levels.[28] Matching was performed considering age at baseline, date of inclusion, sex, smoking habits, and BMI.

Plasma analyses were not available in some of the subjects in the MDC-CC, so analyses were measured in 28 or 34 prevalent patients with IBD and in 106 or 115 controls, respectively. As plasma analyses were not available in several of the few incidental cases, these were omitted from statistical calculations.

All calculations were performed with the SPSS version 22 (Statistical Package for the Social Sciences, IBM Corporation). Calculations were performed separately for the two cohorts compared with controls. Furthermore, the two cohorts were added in an attempt to reach a greater cohort. As age differed between MPM and MDC-CC, and several peptide analyses correlated with age, the values were age-standardized using a linear regression model into which age was added as a covariate and the variables were expressed as z-scores. Those precursors that did not differ between the MPM and MDC-CC cohorts were copeptin ($P = 0.623$), MR-proADM ($P = 0.058$), and MR-proANP ($P = 0.500$), which were thus calculated together. Values are given as median [interquartile range (IQR)] or mean ± standard deviation (SD). The Mann–Whitney *U*-test was used to compare the differences in precursor levels between patients and controls and for subgroup analyses within the IBD group. Fisher's exact test was used for dichotomous variables. The Spearman rank correlation test was used for correlations between precursor and disease duration. $P \leq 0.05$ was considered statistically significant.

Results

Patient characteristics.

MPM. The basal characteristics are described in Table 1. The 22 included patients were in an inactive phase during their enrollment in the study. Disease duration was 25.4 ± 13.4 years. None of the patients was receiving acute therapy, but six (27%) were on continuous treatment with anti-inflammatory or immune-modulating therapy. Twelve patients (54%) had undergone intestinal resections due to IBD, and 10 of the patients (46%) had extensive disease. Apart from IBD, six patients also suffered from hypertension and four from nephrolithiasis. Sporadic cases of asthma bronchialis, bile stones, chronic obstructive pulmonary disease (COPD), dyspepsia, hyperlipidemia, hypothyroidism, migraine, primary Sjögren's syndrome, reflux, and renal insufficiency were found.

MDC-CC. All 40 included patients with prevalent IBD were in an inactive state during enrollment in the study. Disease duration was 13.1 ± 10.0 years (Table 1). None was undergoing acute therapy, and only three patients (8%) were on continuous treatment with anti-inflammatory or immune-modulating therapy. Thirteen patients (32%) had undergone intestinal resections due to IBD, whereas 18 patients (45%) had an extensive distribution of the disease. Sporadic concomitant diseases were present in the form of asthma bronchialis, COPD, duodenal ulceration, dyspepsia, hiatal hernia, hypertension, hypothyroidism, inguinal hernia, ankylosing spondylitis, and venous thrombosis.

Precursor analyses. When precursor analyses were done together, the higher plasma level of copeptin was the only significant difference in plasma measurements between patients and controls (Table 2). Patients who had undergone bowel resection had higher plasma levels of copeptin compared with unresected patients [10.25 (7.94–13.75) and 6.33 (4.92–9.52) pmol/L, respectively, $P = 0.020$]. When the patients with a history of bowel resection were excluded ($n = 23$) together with their matched controls, there was no difference in copeptin levels between patients and controls [6.33 (4.92–9.52) and 5.81 (3.52–9.64) pmol/L, respectively, $P = 0.501$]. There was no dif-

Table 1. Patient characteristics of the two cohorts.

	PREVALENT IBD MPM *n* = 22	PREVALENT IBD MDC-CC *n* = 40	*P*-VALUE
Age (years)	69.7 ± 7.2	56.0 ± 6.5	<0.0001
Sex (male/female)	12/10	19/21	0.791
Smoking (*n*, %)			0.182
Missing values (*n*)		2	
Smokers	11 (50)	12 (30)	
Nonsmokers	11 (50)	26 (65)	
BMI (kg/m^2)	23.8 ± 3.8	24.8 ± 3.5	0.237
Duration of IBD (years)	25.4 ± 13.4	13.1 ± 10.0	<0.0001
Missing values (*n*)		2	
Crohn's disease/ulcerative colitis	8/14	19/21	0.435

Notes: *n* (%) = number and percentage of cases. Values are given as mean ± standard deviation (SD). Mann–Whitney *U*-test or Fisher's exact test. $P \leq 0.05$ was considered statistically significant.
Abbreviations: BMI, body mass index; IBD, inflammatory bowel syndrome; MDC-CC, Malmö Diet and Cancer Study cardiovascular cohort; MPM, Malmö Preventive Medicine.

Table 2. Plasma peptide levels from all patients with inflammatory bowel disease and controls.

	IBD (n = 62)	CONTROLS (n = 186)	P-VALUE
Copeptin (pmol/L)	8.0 (4.8–11.5) (56)	5.8 (3.4–8.5) (181)	0.006
MR-proADM (nmol/L)	0.6 (0.4–0.8) (56)	0.5 (0.4–0.7) (182)	0.127
MR-proANP (pmol/L)	78.0 (59.2–111.8) (56)	72.7 (54.1–101.3) (182)	0.367

Notes: n = number of analyses. Values are given as median [interquartile range (IQR)]. Mann–Whitney U-test. $P \leq 0.05$ was considered statistically significant.
Abbreviations: IBD, inflammatory bowel disease; MR-proADM, midregional fragment of pro-adrenomedullin; MR-proANP, midregional fragment of pro-atrial natriuretic peptide.

ference in copeptin levels whether or not the patients suffered from Crohn's disease or ulcerative colitis [7.19 (4.15–11.34) and 8.13 (5.03–11.68) pmol/L, respectively, P = 0.461], whether they were treated with anti-inflammatory drugs or not [4.69 (3.06–11.77) and 8.09 (5.60–11.43) pmol/L, respectively, P = 0.254], or whether they had a limited or extensive disease [8.03 (5.53–12.60) and 8.13 (5.24–11.05) pmol/L, respectively, P = 0.493].

Since age and disease duration were strongly correlated (rs = 0.374, P = 0.003), the correlation between disease duration and plasma levels of MR-proADM (rs = 0.380, P = 0.005), NT-PTA (rs = 0.483, P = 0.001), and MR-PENK A (rs = 0.302, P = 0.037) was reflected by concomitant correlation with age (rs = 0.668, $P < 0.0001$; rs = 0.483, $P < 0.0001$; and rs = 0.315, P = 0.026, respectively).

The expression of Crohn's disease or ulcerative colitis, treatment with anti-inflammatory drugs or not, limited or extensive disease, or a past bowel resection did not affect the plasma levels of any of the other precursors (data not shown).

When calculated separately, there was a difference between MPM and controls regarding plasma levels of copeptin and MR-proADM but not regarding those of the other precursors (Table 3). The level of copeptin was higher in patients who had undergone a bowel resection (n = 12) compared with the non-resected (n = 6, missing value = 4), although it did not reach statistical significance in the small cohort [11.46 (8.33–19.08) and 7.42 (5.68–14.44), respectively, P = 0.291]. Neither was there any difference in plasma copeptin levels depending on Crohn's disease or ulcerative colitis [7.92 (5.68–11.01) and 10.41 (5.92–16.68), respectively, P = 0.365], were treated with anti-inflammatory drugs or not [9.71 (3.90–12.81) and 8.66 [6.09–18.16] pmol/L, respectively, P = 0.693) or a limited or extensive disease [11.75 (7.08–17.86) and 9.01 (5.90–13.82), respectively, P = 0.604]. Regarding MR-proADM, there was no difference in plasma levels depending on bowel resection or not [0.83 (0.65–1.22) and 0.84 (0.73–1.05), respectively, P = 0.892], Crohn's disease or ulcerative colitis [0.75 (0.69–0.87) and 0.86 (0.66–1.11), respectively, P = 0525], were treated with anti-inflammatory drugs or not [0.84 (0.67–1.11) and 0.79 (0.68–1.01) pmol/L, respectively, P = 0.858], or a limited or extensive disease [0.89 (0.75–1.37) and 0.77 (0.64–1.04), respectively, P = 0.182].

In the MDC-CC cohort, no differences were seen between controls and patients in any plasma precursor analyses (Table 4). Neither were any differences in plasma levels of any of the precursors between the expression of Crohn's disease or ulcerative colitis, treatment with anti-inflammatory drugs or not, limited or extensive disease, or a past bowel resection (data not shown).

Discussion

The main conclusion of this study is that basal plasma copeptin levels are elevated after complete bowel resection, but other peptide precursors are unaffected in middle-aged subjects in inactive IBD compared with matched controls. However, the elevated copeptin levels did not reach statistical significance when precursor levels were calculated separately in the MDC-CC cohort, or when the MPM cohort was separated into

Table 3. Plasma peptide levels from patients in Malmö Preventive Medicine (MPM) compared with controls.

	MPM (n = 22)	CONTROLS (n = 66)	P-VALUE
Copeptin (pmol/L)	8.66 (5.90–12.81)	6.23 (3.79–9.19)	0.007
MR-proADM (nmol/L)	0.80 (0.68–1.04)	0.70 (0.57–0.84)	0.010
MR-proANP (pmol/L)	104.27 (77.10–149.13)	98.73 (68.14–145.69)	0.649
MR-PENK (pmol/L)	59.48 (54.41–74.41)	60.89 (50.39–74.63)	0.785
NT-PTA (pmol/L)	78.98 (64.11–98.66)	80.64 (67.12–97.40)	0.852
Proneurotensin (pmol/L)	79.15 (63.15–130.45)	84.45 (62.18–119.65)	0.972

Notes: n = number of analyses. Values are given as median [interquartile range (IQR)]. Mann–Whitney U-test. $P \leq 0.05$ was considered statistically significant.
Abbreviations: IBD, inflammatory bowel disease; MR-proADM, midregional fragment of pro-adrenomedullin; MR-proANP, midregional fragment of pro-atrial natriuretic peptide; MR-PENK, midregional fragment of proenkephalin A; NT-PTA, N-terminal polytachykinin A.

Table 4. Plasma peptide levels from patients in Malmö Diet and Cancer Study cardiovacsular cohort (MDC-CC) compared with controls.

	MDC-CC (*n* = 40)	CONTROLS (*n* = 120)	*P*-VALUE
Copeptin (pmol/L)	6.70 (3.86–10.55) (34)	5.65 (3.23–8.42) (115)	0.182
MR-proADM (nmol/L)	0.45 (0.38–0.54) (34)	0.45 (0.38–0.51) (116)	0.489
MR-proANP (pmol/L)	66.60 (52.40–82.62) (34)	65.85 (47.38–85.65) (116)	0.499
MR-PENK (pmol/L)	42.65 (37.45–47.52) (28)	46.15 (39.60–53.62) (106)	0.086
NT-PTA (pmol/L)	51.86 (44.18–60.38) (28)	51.01 (41.00–61.67) (102)	0.854
Proneurotensin (pmol/L)	96.28 (62.09–142.69) (28)	93.39 (67.37–138.27) (106)	0.860

Notes: *n* = number of analyses. Values are given as median [interquartile range (IQR)]. Mann–Whitney *U*-test. $P \leq 0.05$ was considered statistically significant.
Abbreviations: IBD, inflammatory bowel disease; MR-proADM, midregional fragment of pro-adrenomedullin; MR-proANP, midregional fragment of pro-atrial natriuretic peptide; MR-PENK, midregional fragment of proenkephalin A; NT-PTA, N-terminal polytachykinin A.

resected and unresected groups. The elevated plasma level of MR-proADM observed in the MPM cohort was diluted when the two cohorts were calculated together.

The etiology of IBD is unknown, but a complex interplay of genetic, microbial, immunologic, and environmental factors are discussed. The inflammation in IBD is mainly situated in the mucosa, but transmural inflammation has been documented in both Crohn's disease and ulcerative colitis.[4] The majority of the neuropeptides are found both in the central nervous system (CNS) and in the ENS, and we do not know whether the plasma peptides are of gastrointestinal or cephalic origin. Gastrointestinal peptides may be secreted from both enterochromaffin mucosal cells and enteric neurons.[9,11,34] Furthermore, IBD affects the brain–gut axis with possible subsequent effects on peptide release from the brain.[35] The modest effect on plasma levels of precursors in the present study suggests that, despite the chronic nature of IBD, the disease does not have a major impact on peptide secretion. Nevertheless, as a wide range of neuropeptides and inflammatory factors are involved in physiological and pathophysiological processes, the balance between peptides, rather than the actual concentration of one specific peptide, may be important.

The levels of copeptin and vasopressin in plasma are correlated with each other in both healthy volunteers and sick patients.[24] Vasopressin has been shown to increase epithelial permeability,[21] but its effect on water reabsorption in the gastrointestinal tract has only been studied in pharmacological doses of vasopressin, and the results are not conclusive.[19,20] As patients with IBD and a history of bowel resection had higher plasma levels of copeptin, the question remains whether the altered precursor levels measured in plasma are a primary etiological factor or a secondary effect of the disease.[7,36] The difference could hypothetically be explained by a compensatory mechanism due to a shorter bowel, with ensuing reduced amount of vasopressin receptors and/or reduced water reabsorption capacity of the bowel.[15,24]

Elevated plasma levels of vasopressin have been found in diseases characterized by chronic inflammation.[37] In mice, injections of vasopressin into the periaqueductal gray raised the plasma levels of all enkephalins,[38] and stimulation of peripheral vasopressin receptors had proinflammatory effects in experimental colitis, whereas inhibition of the receptors abolished the vasopressin effect.[21] The effect of vasopressin on encephalin secretion has not been studied in humans. Proenkephalin is the precursor of Met-enkephalin and Leu-enkephalin, and in contrast to the findings in our study with unaffected MR-PENK A, plasma levels of Met-enkephalin have previously been shown to be lowered in IBD patients.[7,36] The study by Owczarek et al[7] was performed in younger patients with active disease, which could exhaust the peptides in the blood circulation and explain the different results. The actual encephalin concentration in the gastrointestinal tissue is not measured, but agonists to the peripheral opioid receptors lead to anti-inflammatory and anti-nociceptive effects.[13] These studies suggest a role of the enkephalins and their receptors in disease activity. The higher levels of copeptin in IBD patients with a bowel resection may thus be of interest, as vasopressin may modulate gastrointestinal inflammation, either directly[38] or indirectly through modulation of proenkephalins and their stimulation of the opioid receptors.[13,21] The role of copeptin to keep the IBD patient in remission deserves to be further examined.

IBS-like symptoms are present in 35%–40% of IBD patients during remission. This well-described phenomenon has for several years been believed to depend on low-grade inflammation.[3] Very few human studies have examined the effect of neuropeptides on visceral pain, but a connection between spinal afferents, enteric mast cells, and the ENS has been confirmed, suggesting how elevation of intestinal hypersensitivity might occur involving neural peptides, eg, neurotensin and substance P.[12,14] Substance P has also been shown to exert proinflammatory effects, especially on neurogenic inflammation.[11,14] Noninflamed small bowel samples from patients with Crohn's disease have shown increased expression of substance P mRNA in one study,[5] with more binding sites in the inflamed areas, although unchanged mRNA levels were found in another study.[6] Plasma levels of substance P have previously been found to be increased in IBS patients but not in IBD patients with IBS-like symptoms.[39] Presence of IBS-like symptoms was not measured in this retrospective study, but the unaltered plasma levels of precursors in the current study do

not suggest involvement of peptides for development of gastrointestinal symptoms.

The limitation of the present study is the small study population. However, previous studies in the field have been of smaller or similar sizes.[7,36] The advantage of measuring precursors instead of peptides depends on the high stability of these precursors, which are synthesized in stoichiometric amounts relative to the mature peptides.[23,24] The strength is that we have analyzed several precursors in well-characterized cohorts. Higher plasma levels of precursors may be related to a reduced degradation/excretion in an elder cohort, rather than an altered expression/secretion.[40] This means that multiple factors influence the precursor levels in plasma. We have matched for age, gender, smoking habits, and BMI, and excluded patients with cardiovascular diseases, diabetes mellitus, and malignancy, but factors such as dietary habits, alcohol intake, stress, physical activity, sleeping habits, and drug therapy may also influence the secretion and degradation of precursors. The difference between the two cohorts regarding plasma values of PENK, PNT, and PTA may be reflected by the different factors mentioned above. Although not statistically significant, there were more cases of Crohn's disease in the MDC-CC cohort than in the MPM cohort, and also a different smoking prevalence between the two cohorts. As data on the MPM cohort were collected several years before those of the MDC-CC cohort, treatment regimens in the daily clinic also differed, which may have influenced the precursor secretion. Thus, the two cohorts were not matched or identical in composition.

In conclusion, higher basal plasma levels of copeptin were found in inactive IBD patients, whereas the basal plasma levels of MR-proADM, MR-proANP, MR-PENK A, NT-PTA, and proneurotensin were not altered in inactive IBD compared with controls in middle-aged subjects. Elevated copeptin levels were found in IBD patients who had undergone bowel resection. The role of copeptin to keep the IBD patient in remission deserves to be further examined.

List of Abbreviations

AVP, arginine vasopressin; BMI, body mass index; IBD, inflammatory bowel disease; MDC-CC, Malmö Diet and Cancer Study cardiovascular cohort; MPM, Malmö Preventive Medicine; MR-proADM, midregional fragment of pro-adrenomedullin; MR-proANP, midregional fragment of pro-atrial natriuretic peptide; MR-PENK, midregional fragment of proenkephalin A; NT-PTA, n-terminal polytachykinin A.

Author Contributions

Made substantial contributions to the conception and design of the study, participated in the interpretation of the statistical analysis, and financed the study: BO, OM. Validated the medical records and wrote the manuscript: BO. Both authors were involved in revising the manuscript critically for important intellectual content, and approved the final manuscript.

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