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ORIGINAL ARTICLE

Association of the dopamine D2 receptor gene SNP rs1800497 with postoperative nausea and vomiting

A prospective cohort study

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BACKGROUND Postoperative nausea and vomiting (PONV) are the most frequent complications in the context of anaesthesia. Several studies suggest a contribution of genetic traits to PONV disposition. Single nucleotide polymorphisms (SNPs) located in the cholinergic receptor muscarinic 3 gene *CHRM3* (rs2165870) and the potassium voltage-gated channel subfamily B member 2 *KCNB2* (rs349358) have been described as independent risk factors for the occurrence of PONV. In addition, further SNPs might be associated with an increased PONV risk, for example a dopamine D2 receptor (*DRD2*) SNP (rs1800497).

OBJECTIVE The primary aim of our study was the development of a new PONV prediction score which includes genetic information of SNPs in the genes *CHRM3* and *KCNB2,* which have been already associated with PONV. The secondary aim of our study was to investigate the association of five additional SNPs with PONV.

DESIGN Prospective cohort study.

SETTING Single centre study in Germany.

RESULTS We could not establish a new PONV prediction score that includes genetic information, due to limited association of the *KCNB2* SNP and *CHRM3* SNP with PONV. Interestingly, the GA and AA genotypes of the *DRD2* rs1800497 in the dopamine D2 receptor gene were associated with PONV 24 h postoperatively, with a relative risk (RR) of GA/AA genotype vs. GG genotype of 1.5 [95% confidence interval (CI) 1.06 to 2.01, $P = 0.02$]. This association was independent from the Apfel score in a multivariate logistic regression analysis (RR 1.4, 95% CI 1.03 to 1.90, $P = 0.03$).

CONCLUSION The construction of a new PONV prediction score including genetic information was not possible due to limited association of the *CHRM3* and *KCNB2* SNPs. However, the *DRD2* GA and AA genotypes (rs1800497) were associated with PONV and this SNP might be a future candidate for further validation studies aiming for molecular-derived PONV prediction models.

TRIAL REGISTRATION German Clinical Study Register – DRKS00021051.

Introduction

Postoperative nausea and vomiting (PONV) are the most common adverse effects of general anaesthesia with a prevalence of 20 to 30% in unselected patient cohorts.[1,2] In an outcome study, the majority of patients ranked PONV as the most unpleasant adverse event after surgery, rated as even more undesirable than postoperative pain.[3] In a survey, patients were willing to pay 56 to 100 USD to prevent PONV.[4] Therefore, avoidance of PONV is a quality criterion for anaesthesia and improves perioperative satisfaction. Moreover, extensive postoperative

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vomiting can result in further complications such as aspiration, impaired wound healing, electrolyte imbalance, Boerhaave's syndrome or even pneumothoraces or subcutaneous emphysema.[5–7]

In 1999, Apfel *et al.* described a risk score that is now a commonly used tool to predict PONV in adults.[8] It consists of the four clinical risk factors female gender, non-smoking status, previous history of PONV or motion sickness, and anticipated need for postoperative opioids. According to this score, occurrence of no risk factor results in a relative PONV risk of 10%, one risk factor, 21%, two risk factors, 39%, three risk factors, 61%, and four risk factors, 79%.[8]

The current international guideline for the management of PONV recommends a multimodal approach for PONV prevention, with prophylaxis by antiemetic drugs and use of total intravenous anaesthesia (TIVA) being important strategies.[1] Each antiemetic prophylactic reduces the risk for PONV by about 26%, and the effect of a combination of antiemetics is additive.[9] Often, a risk-adapted algorithm for prophylaxis is applied based on the risk determined by the Apfel score.[1]

However, several genetic factors contributing to PONV risk have been identified. In a genome-wide association study (GWAS) a single nucleotide polymorphism (SNP) upstream of the muscarinic acetylcholine receptor 3 subtype (*CHRM3*) gene (rs2165870) was associated with PONV[10] and this association was confirmed by two independent studies.[11,12] Also, a SNP in the voltage-gated potassium channel subfamily B member 2 (*KCNB2*) gene (rs349358) was associated with PONV in the GWAS and this was confirmed in a further study cohort.[11,13]

Additionally, in several hypothesis-driven studies further SNPs were associated with PONV.[13,14] The most promising candidates were SNPs in relation to the genes of the μ-opioid receptor (*OPRMI*) rs179997,[15] the dopamine D2 receptor (*DRD2*) rs1800497,[16,17] the solute carrier family 6 A 4 denoted as 5 hydroxytrypamine transporter linked promoter region (*5-HTTLPR*) rs4795541,[18,19] the 5 hydroxytrypamine receptor 3A (*HTR3A)* rs1176713[20] and the interleukin 2 receptor type B (*ILR2B*) rs3218315.[10,13]

Taking this into account, the traditional approach for PONV prophylaxis strictly according to the Apfel score might be insufficient to detect all patients susceptible for PONV. Confounding factors caused by genetic traits might be neglected. Therefore, the primary aim of our study was to develop a combined risk stratification score for PONV including contributing genetic factors (the *CHRM3* SNP rs2165870 and *KCNB2* SNP rs348358). The secondary aim was to investigate putative associations of five SNPs already described in the context of PONV in our study cohort, including the *DRD2* SNP (rs1800497).

Methods

Study procedures

This prospective single centre study investigating the prediction of PONV based on genetic variables was approved by the Ethics Committee of the University of Duisburg-Essen, Medical Faculty (study identifier 19-8880-BO) on 09/03/2020. All investigations were conducted in accordance with the latest version of the Declaration of Helsinki and the STROBE statement for observational studies. The study was registered in the German Clinical Study Register (DRKS00021051). We conducted the study at the University Hospital of Essen. After obtaining written informed consent, patients of either sex older than 18 years of age, scheduled to undergo general anaesthesia for elective surgical procedures in the departments of otorhinolaryngology, gynaecology, urology, ophthalmology, spinal surgery, orthopaedic surgery, and visceral surgery were eligible for inclusion in the study. Patients requiring neurosurgical head interventions or cardiac surgery were excluded, as well as patients managed with prolonged postoperative sedation and mechanical ventilation in the intensive care unit (ICU).

Anaesthesia

All patients received a standardised anaesthesia regimen including induction of anaesthesia with either propofol (1 to 3 mg kg^{-1}), thiopentone (5 mg kg^{-1}), or etomidate (0.3 mg kg^{-1}), and maintenance of anaesthesia using volatile anaesthetics (isoflurane in a minimal alveolar concentration (MAC) of 0.4 to 1.6; sevoflurane in a MAC of 0.8 to 3.7, or desflurane in a MAC 2.1 to 8.8) in oxygen/air or optional oxygen/nitrous oxide. Intraoperative analgesia consisted of fentanyl in a dose of 0.3 μg kg^{-1}. Patients receiving regional anaesthesia, including single shot procedures, were excluded. Postanaesthetic analgesia was achieved using piritramide or morphine intravenously in a dose of 0.1 mg kg^{-1}. Gastric tubes were removed during extubation as standard.

Postoperative nausea and vomiting prophylaxis

Because this investigation was designed as an observational study, the PONV prophylaxis regimen was chosen by the anaesthesiologist responsible and did not follow a strict study regimen. Prophylactic agents used were the 5-HT3$_A$-receptor antagonist granisetron (1 to 3 mg), glucocorticoids in the form of dexamethasone (4 mg) and methylprednisolone (40 to 125 mg), and/or dopamine receptor antagonists (haloperidol 1 mg). In case of severe PONV in the post-anaesthetic care unit (PACU), patients received an antiemetic rescue of dimenhydrinate (62 mg in an i.v. infusion over a period of 15 min) or granisetron (1 to 3 mg), if not administered prophylactically.

Measurements of clinical outcome

The occurrence of postoperative nausea, vomiting and retching, and also pain, was recorded using a structured survey form categorising pain and nausea according to the

visual analogous scale from 0 (no complaints) to 10 (major impairment/severe sensation of pain or nausea). Nausea was defined as sickness without the need to retch or vomit, and retching was defined as a salvo of delayed vomiting in multiple gradings in contrast to the act of vomiting per se. To correct for a constant condition of sickness or a preoperative existing urge to vomit, patients were asked for their level of nausea prior to induction of anaesthesia. Simultaneously, recovery from anaesthesia and satisfaction with antiemetic therapy were surveyed on a range from 0 (major impairment/insufficient recovery) to 100 (no complaints/full recovery). The form was filled in by medical staff after interviewing the patients directly postoperatively (in the PACU), in the early postoperative period (2 to 6 h), and in the late postoperative period (6 to 24 h).

Genotyping

DNA was extracted from buccal mouth swabs prior to the elective surgery using the QIAamp DNA blood mini kit (Qiagen, Venlo, Netherlands) according to the manufacturer's recommendations. Genotyping was carried out using VIC/FAM labelled TaqMan SNP genotyping assay (Thermo Fisher Scientific, Waltham, MA, USA) in a StepOnePlus real time polymerase chain reaction (PCR) system (Applied Biosystems, Waltham, MA, USA). 5-HTTLPR (rs4795541) was investigated by PCR amplification and agarose gel electrophoresis with primers 5-HTTLPR_for 5'-GGCGTTGCCGCTCTGAATGC-3' and 5-HTTLPR_rev 5'-GAGGGACTGAGCTGGACAACCAC-3'. Short and long alleles differed by 43 bp in length (short product: 486 bp in length, long product: 529 bp in length).

Statistical analysis

Personal data are depicted as frequencies (%) or median [range] as appropriate. We assessed the Gaussian distribution of data by using the Kolmogorov-Smirnov normality test. In case of nonparametric data distribution, Mann–Whitney *U*-test and Kruskal–Wallis tests were applied. Calculation of the genotype distribution according to the Hardy Weinberg (HW) equilibrium was conducted using the HW calculator (M. H. Court, 2005 to 2008, Court MH. Court-laboratory Hardy-Weinberg calculator. 2005 to 2008. http://www.tufts.edu/~mcourt01/Documents/Court%20lab%20%20HW%20calculator.xls). Categorical variables were compared using cross tables (χ^2 test). We used binary logistic regression models to calculate the impact of individual variables for the risk of postoperative nausea and vomiting, 95% confidence intervals and *P* values. Pre-described risk factors for PONV were included in multivariate logistic regression analysis, including duration of anaesthesia, age, intra-operative use of nitrous oxide, risk surgery defined as cholecystectomy, gynaecology, or laparoscopic surgery, and the factors of the Apfel score (history of PONV or motion sickness, female sex, postoperative need for opioids, non-smoking status). Binominal confidence intervals were calculated using the Clopper Pearson interval. The study was powered for the primary study aim to develop a risk score based on previous study results of the *KCNB2* and *CHRM3* genotypes in addition to known clinical variables of PONV risk (female sex, history of PONV and/or motion sickness, non-smoking status, age, duration of anaesthesia, postoperative need for opioids). The power calculation of these 10 variables resulted in a minimal need for $n = 700$ patients, aiming for a statistical probability of at least seven events per criterion in our study population. To reach a reasonable amount of statistical safety, the minimal target number of included patients was extended to $n = 800$ patients. We produced graphs using GraphPad Prism Software version 8.1.2 (Boston, MA, USA). Statistical analysis was performed using SPSS Statistics version 27 (IBM, Armonk, NY, USA). Alpha errors *P* of <0.05 were considered as statistically significant.

Results

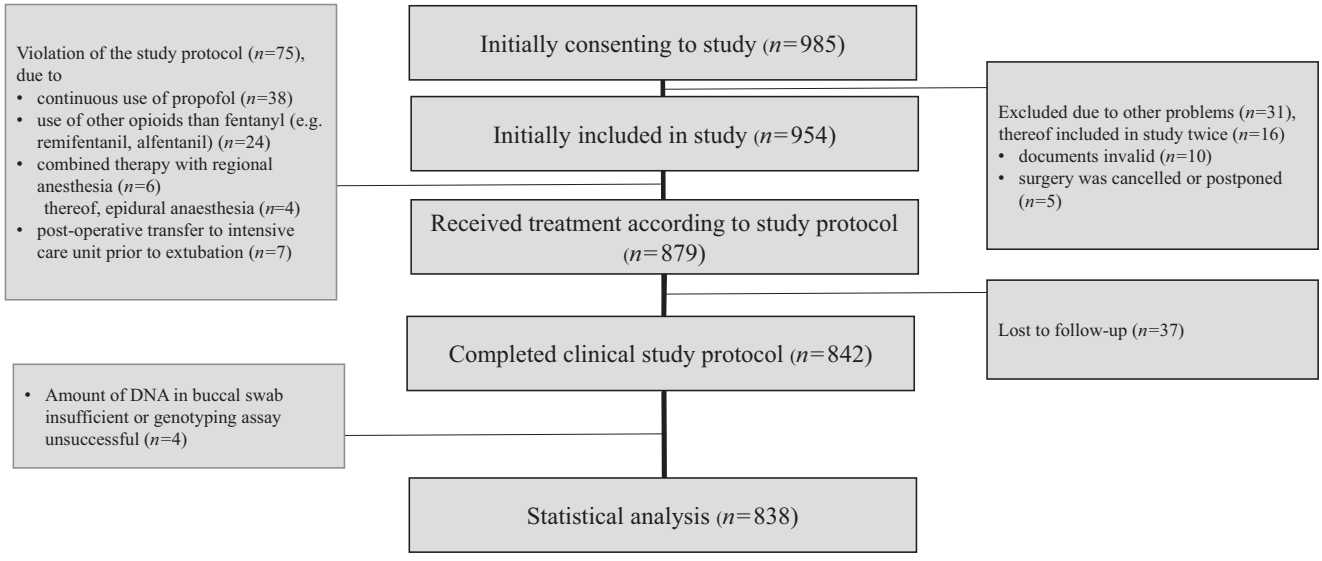
Patients were recruited from September 2020 to September 2022. The study period included the SARS CoV2 pandemic, leading to a reduction in the total amount of electively scheduled surgical procedures. The number of patients eligible to our study was a priori limited due to tight exclusion criteria (total intravenous anaesthesia, additional regional anaesthetic procedures, use of other opioids than fentanyl, and others). In total, 1254 patients were assessed for eligibility, of which 269 patients refused to participate (Fig. 1), 985 initially consented to the study, 879 received treatment according to the study protocol and 37 were lost to follow up leaving 842 who completed the clinical study protocol. For four of these, an insufficient amount of DNA was obtained, leaving 838 for statistical analysis (Fig. 1, Table 1).

Minor allele frequencies (MAF) of all initially included individuals ($n = 966$) were compared to genotyping data of an open access genome database (gnomAD, Broad Institute, Cambridge, MA, USA),[21] which uses inter alia underlying data from the 1000 genomes project.[22] In our study, MAFs did not significantly deviate from data of a representative European non-Finnish population, and genotype distributions did not significantly deviate from the Hardy Weinberg equilibrium.

Occurrence of postoperative nausea and vomiting

Overall, PONV was detectable in 34% of the patients investigated, of which 33.8% experienced postoperative nausea. Most frequently, nausea occurred in the early phase of 2 to 6 postoperative hours, followed by 18% who experienced PONV immediately in the PACU. In 6%, PONV occurred in the late period 6 to 24 h postoperatively (Fig. 2). Overall, 15.8% of patients in our study cohort suffered from retching and/or vomiting. Here again, the most vulnerable time frame was within the early phase (2 to 6 h postoperatively), when retching and/or vomiting occurred in 10% of patients (Fig. 2). In 8%,

FIGURE 1 Study flow chart.



retching and/or vomiting began in the PACU and in 1% in the late phase (6 to 24 h).

Development of a new postoperative nausea and vomiting prediction score

Although sufficiently powered, we could not find statistically significant associations of PONV, nausea or retching/vomiting with genotypes of the SNP *KCNB2* (rs34958) within the time frame of the first 24 postoperative hours (PONV overall TT genotype 33.6%, TC genotype 34.5%, CC genotype 41.7%, $P = 0.63$; nausea: TT genotype 33.2%, TC genotype 34.5%, CC genotype 41.7%, $P = 0.61$; retching/vomiting: TT genotype 15.1%, TC genotype 16.5%, CC genotype 25%, $P = 0.53$; Table 2).

χ^2-analysis of the *CHRM3* SNP (rs2165870) just achieved a significant association of the GG genotype with overall PONV (GG genotype 37.6%, GA genotype 33.0%, AA genotype 25.4%, $P = 0.05$), which was also discernible for nausea (GG genotype 37.3%, GA genotype 32.7%, AA genotype 25.4%, $P = 0.05$). There was no association for retching/vomiting (GG genotype 17.6%, GA genotype 14.9%, AA genotype 12.3%, $P = 0.33$) in the first 24 h (Table 2). Due to this limited association, the elaboration of a new PONV prediction score based on *KCNB2* and *CHRM3* genotype patterns was not meaningful.

Association of further single nucleotide polymorphisms with postoperative nausea and vomiting

Interestingly, the *DRD2* GA/AA genotype (rs1800497) was significantly associated with PONV 24 h postoperatively (GA/AA genotype 39.1%, GG genotype 31.1%, $P = 0.02$), which was accompanied by a significant association of the GA/AA genotype with nausea within the first 24 h (GA/AA genotype 39.1%, GG genotype 30.7%, $P = 0.01$).

HTR3A (rs1176713), *OPRM1* (rs1799971), *IL2RB* (rs3218315), and *5-HTTLPR* (rs4795541) were not associated with PONV (see Table S1, Supplemental Digital Content, http://links.lww.com/EJAIC/A76). Therefore, we further investigated the *DRD2* SNP (rs1800497) in secondary analyses.

Dopamine D2 receptor single nucleotide polymorphism rs1800497

As described in the first *DRD2* (rs1800497) association study, the SNP was assessed in a dominant model combining the minor AA genotype and heterozygous patients.[16] In a univariate pairwise analysis, the combination of AA and GA genotype led to an increase in relative PONV risk of about 42.3% in comparison to the major GG genotype ($P = 0.02$). We compared the *DRD2* GA/AA genotype with the GG genotype concerning the personal variables age, height, weight, sex, history of PONV, motion sickness, and smoking, duration of narcosis, ASA score, Apfel score, and amount of fentanyl used intra-operatively. No significant differences were detected within the genotype groups (Table 3). Therefore, differences in PONV susceptibility were not attributable to intervariation within the study cohort.

Considering further known risk factors contributing to PONV, we subjected the *DRD2* SNP to a multivariate regression analysis (Table 4). Here, it proved an independent risk factor for PONV ($P = 0.02$, RR 1.46 in comparison to GG genotype, 95% CI 1.06 to 2.01). Simultaneously, multiple known risk factors for PONV, such as the variables of the Apfel score, were confirmed in our study cohort. Of these, female sex ($P < 0.001$, RR 2.01, 95% CI 1.42 to 2.86), history of PONV and/or

Table 1 Personal and clinical data ($n = 838$) given as *n* (%) or median [range] as appropriate

Characteristic	Occurrence
Female/male	476 (56.8)/362 (43.2)
Weight, kg	80 [38 to 194]
Height, cm	172 [150 to 203]
BMI, kg m^{-2}	26.27 [13.15 to 56.46]
Age, years	56 [18 to 89]
Apfel score	
0–1	251 (30.0)
2	261 (31.1)
3	239 (28.5)
4	87 (10.4)
Smoking status	
current smoker	238 (28.4)
ex (*defined as non-smoker < 12 months*)	33 (3.9)
non-smoker	567 (67.7)
Surgical department	
visceral surgery	181 (21.6)
gynaecology	174 (20.8)
urology	163 (19.5)
orthopaedic surgery	149 (17.8)
neurosurgery	129 (15.4)
otorhinolaryngology	42 (5.0)
ASA classification	
1	81 (9.7)
2	515 (61.5)
3	239 (28.5)
4	3 (0.4)
Risk surgery	270 (32.2)
includes laparoscopy, gynaecology, CHE	
Intra-operative use of N_2O	179 (21.4)
Dose of intra-operative fentanyl, mg	0.25 [0.05 to 2]
Induction	
propofol	821 (98)
etomidate	11 (1.3)
thiopentone	9 (1.1)
Maintenance	
sevoflurane	753 (89.9)
isoflurane	19 (2.3)
desflurane	67 (8.0)
Patients receiving PONV prophylaxis	478 (57.0)
Medication for PONV prophylaxis	
no prophylaxis	360 (43.0)
1 drug	334 (39.9)
2 drugs	143 (17.1)
3 drugs	1 (0.1)
Patients receiving PONV prophylaxis using	
dexamethasone	435 (51.9)
granisetron	184 (22)
haloperidol	2 (0.2)

ASA, American society of Anesthesiologists; BMI, body mass index. CHE: cholecystectomy.

motion sickness ($P < 0.001$, RR 2.63, 95% CI 1.84 to 3.78), and postoperative need for opioids ($P = 0.03$, RR 1.47, 95% CI 1.05 to 2.07) were significantly associated with PONV within 24 h (Table 4). Furthermore, the intra-operative use of nitrous oxide ($P = 0.03$, RR 1.52, 95% CI 1.05 to 2.20) and the conduction of risk surgery ($P = 0.001$, RR 1.77, 95% CI 1.25 to 2.49) were significantly associated. In our study cohort there was no detectable significant association of non-smoking status ($P = 0.50$) and duration of anaesthesia ($P = 0.13$) (Table 4).

To assess whether the significant association found for the *DRD2* SNP was independent of the Apfel score, a multivariate logistic regression analysis with the *DRD2* SNP and the Apfel score was carried out. Here, the *DRD2* GA/AA genotypes were identified as independent risk factors for PONV over 24 h apart from the Apfel score ($P = 0.03$). The GA/AA genotype contributed to a relative PONV risk of 39.7% (95% CI 1.03 to 1.9), whereas the cumulative consideration of the variables of the well established Apfel score resulted in a relative PONV risk of 1.92 per score point in our study cohort (95% CI 1.65 to 2.23).

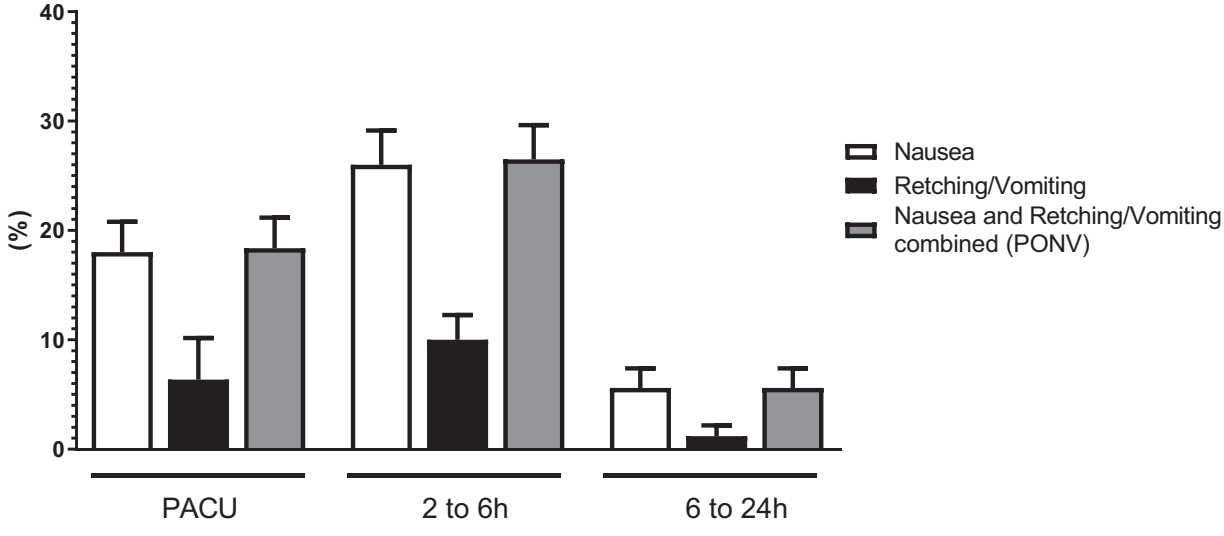
Discussion

The primary aim of our study was to develop a new PONV prediction score using pre-described associations of *CHRM3* SNP rs2165870 and *KCNB2* SNP rs348358 with PONV in combination with clinical variables. In our study cohort, however, we found a limited association of the *CHRM3* rs2165870 and the *KCNB2* rs348358 with PONV (Table 2). Remarkably, the *CHRM3* rs2165870 GG genotype was marginally significantly associated with PONV postoperatively ($P = 0.05$), whereas in previous studies the AA genotype of the *CHRM3* rs2165870 SNP was associated with PONV.[21] Unfortunately, the development of a new PONV prediction score with consideration of genetic and clinical characteristics was therefore not meaningful.

We had been led to our primary study objective by a strong and replicated association of the *CHRM3* rs2165870 and the *KCNB2* rs348358 with PONV.[10–13] Therefore, we compared our current with previous studies to reach possible explanations for the divergent results. Particular attention was paid to the ethnicity and size of study cohort, anaesthetic regimens used, study modalities, and their main findings (Table 5).

In a GWAS study the *CHRM3* rs2165870 and *KCNB2* rs349358 were significantly associated with PONV.[10] In a verification study, our group found a significant association of the rs2165870 AA genotype with the occurrence of PONV within 2 to 6 h in a Caucasian population, which was further confirmed in a Han Chinese cohort.[11,12] Another study investigating a Caucasian cohort, however, failed to reproduce an association with PONV neither within the first 24 h, nor within 48 h postoperatively.[23] A review speculated that different anaesthesia regimens and different ethnicities of participants contributed to heterogenic findings.[14] Our mini-review concerning these SNPs also underlines differences in study modalities, especially concerning the maintenance of anaesthesia using different volatile anaesthetics or even continuous propofol infusion (Table 5). In our previous study, isoflurane in combination with nitrous oxide was used.[11] In this study however, a high percentage of patients received sevoflurane, whereas the continuous use of propofol was an exclusion criterion, and nitrous oxide supplemented volatile anaesthetics in only 21.4% of all cases evaluated (Table 1). In the Han Chinese

FIGURE 2 Occurrence of nausea, retching/vomiting and combined PONV within the postoperative periods investigated. Depicted are means in percentages and 95% confidence intervals. Calculation of binominal confidence intervals using the Clopper Pearson interval. PACU, post-anaesthetic care unit.



patient cohort, Wang *et al.* used continuous propofol infusions in combination with sevoflurane to maintain anaesthesia.[12] In a Japanese investigation, general anaesthesia was combined with epidural techniques.[24] Since the technique of anaesthesia and medication used for anaesthesia differed in all studies, the comparability of the results is limited (Table 5). Differences in the major allele frequencies (MAF) in different ethnicities also have to be taken into account (*CHRM3* rs2165870 MAF Caucasian: 0.67, MAF East-Asian: 0.85, MAF African: 0.93).

The secondary aim of our study was to investigate the association of five hypothesis-driven SNPs with the occurrence of PONV in our study cohort. In most SNPs investigated, namely *HTR3A* (rs1176713), *OPRM1* (rs1799971), *IL2RB* (rs3218315), and *5-HTTLPR* (rs4795541), we could not reveal a significant association with the occurrence of PONV within 24 h (see Table S2, Supplemental Digital Content, http://links.lww.com/EJAIC/A76).

However, we did show an independent association of the rs1800497 SNP in close linkage disequilibrium with the dopamine D2 receptor gene (*DRD2*) with the incidence of PONV within the first 24 h ($P = 0.02$). The *DRD2* GA/AA genotype was independently associated with PONV ($P = 0.02$) in a multivariate analysis, in concordance with other well known factors contributing to PONV (female sex, history of PONV or motion sickness, non-smoking status, postoperative need for opioids, age, intra-operative use of nitrous oxide, risk surgery, Table 4). More

Table 2 **Association of the *CHRM3* and *KCNB2* genotypes with PONV, nausea, and retching/vomiting over 24 h**

KCNB2	**Total**	**TT genotype**	**TC genotype**	**CC genotype**	***P* value**
No PONV	553 (66)	366 (66.4)	171 (65.5)	14 (58.3)	0.63
PONV	285 (34)	185 (33.6)	90 (34.5)	10 (41.7)	
No nausea	555 (66.2)	368 (66.8)	171 (65.5)	14 (58.3)	0.61
Nausea	283 (33.8)	186 (33.2)	90 (34.5)	10 (41.7)	
No vomiting/retching	706 (84.2)	468 (84.9)	218 (83.5)	18 (75)	0.53
Vomiting/retching	132 (15.8)	83 (15.1)	43 (16.5)	6 (25)	
CHRM3	**Total**	**GG genotype**	**GA genotype**	**AA genotype**	***P* value**
No PONV	553 (66)	234 (62.4)	234 (67)	85 (74.6)	0.05
PONV	285 (34)	141 (37.6)	115 (33)	29 (25.4)	
No nausea	555 (66.2)	235 (62.7)	235 (67.3)	85 (74.6)	0.05
Nausea	283 (33.8)	140 (37.3)	114 (32.7)	29 (25.4)	
No vomiting/retching	706 (84.2)	309 (82.4)	297 (85.1)	100 (87.7)	0.33
Vomiting/retching	132 (15.8)	66 (17.6)	52 (14.9)	14 (12.3)	

Data are given as *n* (%). χ^2-calculation of two-tailed asymptotic significance. A, adenine; C, cytosine; *CHRM3*, muscarinic acetylcholine receptor 3 subtype; G, guanine; *KCNB2*, voltage-gated potassium channel subfamiliy B member 2; T, thymidine.

Table 3 Intervariate risk stratification of genotype groups of the *DRD2* SNP

Variable	Total	GA/AA genotype	GG genotype	*P* value
N	838	307	531	
Age, years	56 [18 to 89]	54 [19 to 88]	57 [8 to 89]	0.15
Height, cm	172 [150 to 203]	172 [150 to 200]	172 [150 to 203)]	0.48
Weight, kg	80 [38 to 194]	80 [38 to 183]	79 [40 to 194]	0.80
Sex (f/m)	476/362	180/127	296/235	0.42
Smoker	259 (30.9)	93 (30.3)	166 (31.3)	0.77
History of motion sickness	102 (12.2)	39 (12.7)	63 (11.9)	0.72
History of PONV	128 (15.3)	48 (15.6)	80 (15.1)	0.83
Duration of narcosis, min	135 [20 to 510]	130 [20 to 510]	140 [25 to 472]	0.16
ASA score				
1	81 (9.7)	35 (11.4)	46 (8.7)	0.19
2	515 (61.5)	189 (61.6)	326 (61.4)	
3	239 (28.5)	83 (27)	156 (29.4)	
4	3 (0.4)	0 (0)	3 (0.6)	
Amount of intra-operative fentanyl, mg	0.25 [0.05 to 2.0]	0.25 [0.1 to 1.4]	0.25 [0.05 to 2.0]	0.19
Apfel score				
0–1	251 (30)	86 (28)	165 (31.1)	1.27
2	261 (31.1)	97 (31.6)	164 (30.9)	
3	239 (28.5)	91 (29.6)	148 (27.9)	
4	87 (10.4)	33 (10.7)	54 (10.2)	

Data are given as number, *n* (%) or median [range] as appropriate. No significant differences were found within the genotype groups. Nonparametric data according to Kolmogorov–Smirnov test for normality. *P* values are calculated using the Mann–Whitney *U*-test or Kruskal–Wallis test accordingly. A, adenine; ASA, American Society for Anesthesiologists; G, guanine.

extensively, this association was independent of the Apfel score ($P = 0.03$).

rs1800497 is referred to as a *DRD2* SNP, but originally corresponds to a SNP in the ankyrin repeat and kinase domain containing 1 (*ANKK1*) gene, which is located 10 000 bp downstream of *DRD2* on chromosome 11q23.2.[25–27] The polymorphism is based on a missense mutation G > A on the plus strand of the gene, resulting in an amino acid exchange from glutamine to lysine in the ANKK1 protein.[25] Antagonists on dopamine D2 receptor (DRD2), such as droperidol, amisulpride or metoclopramide, are frequently used drugs for antiemetic therapy.[1] Droperidol is established as an efficient prophylactic drug for PONV in international consensus guidelines.[1] The initial investigations of *DRD2* SNP associations with PONV arose from hypotheses generated upon this pathophysiological connection between DRD2-antagonism and PONV relief.

Regarding *DRD2* rs1800497 in the context of postoperative vomiting, the GG genotype was described as an independent risk factor for developing PONV in a dominant genetic model, investigated in a Japanese study cohort within 6 h after surgery.[16] This was further elucidated in a study with patients of Caucasian origin.[17] Although here the GG genotype was associated with a history of PONV, the *DRD2* rs1800497 SNP could not be identified as an independent risk factor for PONV.[17]

In our study, the GA and AA genotype contributed to an increased PONV risk. From a detailed overview of previous publications we found a number of differences in study design and cohort characteristics (Table 6). Nakagawa *et al*. conducted their study in a Japanese cohort, and this difference in ethnicity is important. In the Japanese, the minor allele frequency of *DRD2* rs1800497 deviates considerably (A = 0.37) from the European minor allele frequency (A = 0.19), which might be a reason for differing results. Furthermore, all studies, whether they found significant associations or not, showed specific differences in anaesthetic regimens, characteristics of the study cohort, and in the study concept (Table 6). Furthermore, it is uncertain if the SNP investigated in our study (rs1800497) is the only polymorphism within this gene responsible for the association with PONV. Other SNPs

Table 4 Binary logistic regression analysis of *DRD2* SNP in comparison to further approved risk factors for PONV

Variable	Relative risk	95% confidence interval	*P* value
GA/AA genotype	1.46	1.06 to 2.01	0.02
Female sex	2.01	1.42 to 2.86	<0.001
History of PONV and/or motion sickness	2.63	1.84 to 3.78	<0.001
Non-smoking status	1.13	0.80 to 1.59	0.5
Postoperative need for opioids	1.47	1.05 to 2.07	0.03
Duration of anaesthesia	1.002	1.00 to 1.004	0.13
Age	0.99	0.98 to 0.1	0.01
Intra-operative use of nitrous oxide	1.52	1.05 to 2.2	0.03
Risk surgery	1.77	1.25 to 2.49	0.001

Statistics: binary logistic regression analysis. A, adenine; G, guanine.

Table 5 Comparison of different study results investigating the association of *CHRM3* rs2165870 and *KCNB2* rs349358 with PONV

SNP	Authors	Study cohort	Ethnicity	Anaesthesia	Main findings
CHRM3 (rs2165870) *KCNB2* (rs349358)	Janicki *et al.* 2011[10]	$n=251$ ($n=122$ PONV $n=129$ control)	Caucasian	- general anaesthesia - maintenance: volatile anaesthetics	- GWAS identified *CHRM3* rs2165870 as significantly associated with PONV risk, which acts as highest-ranking SNP associated with PONV risk after correction for multiple testing ($P=0.000055$). - *KCNB2* (rs349358) is significantly associated with PONV risk ($P=0.048$).
CHRM3 (rs2165870)	Hayashi *et al.*, 2012[24]	$n=70$ (women only)	Japanese	- general anaesthesia in combination with epidural anaesthesia - gynaecological surgery only - induction: propofol and fentanyl - maintenance: sevoflurane in oxygen	- Need for antiemetics was more frequent in patients with AA genotype in comparison to patients with the GG genotype ($P=0.008$).
CHRM3 (rs2165870)	Klenke *et al.* 2018[11]	$n=454$	European	- general anaesthesia - induction: thiopentone, propofol or etomidate, fentanyl, and rocuronium or succinylcholine - maintenance: isoflurane + nitrous oxide	- *CHRM3* rs2165870 homozygous AA genotypes significantly contributed to nausea, retching and vomiting 2 to 6 h postoperatively (30% greater PONV risk in AA homozygous genotypes, $P=0.005$). - *CHRM3* rs2165870 AA genotype independently contributed to PONV risk apart from the Apfel score.
CHRM3 (rs2165870)	Wang *et al.*, 2020[12]	$n=512$	Han Chinese	- general anaesthesia - induction: fentanyl, midazolam, etomidate, and vecuronium bromide - maintenance: continuous propofol infusion (4 mg kg^{-1} h^{-1}) + sevoflurane (MAC 1to2)	- *CHRM3* rs2165870 AA genotype was related to an increased PONV risk ($P=0.001$) within 2 to 6 h. - *CHRM3* rs2165870 AA genotype was associated with a higher incidence of PONV in patients receiving ondansetron 0 to 2 h postoperatively.
CHRM3 (rs2165870)	Gloor *et al.*, 2021[23]	$n=613$	European	- anaesthesia not further specified - maintenance: continuous propofol infusion or volatile anaesthetics	- *CHRM3* rs2165870: no significant association with PONV was detected within the first 24 and 48 h.
KCNB2 (rs349358)	Klenke *et al.* 2018[11]	$n=465$	European	- general anaesthesia - induction: thiopentone, propofol or etomidate, fentanyl, and rocuronium or succinylcholine - maintenance: isoflurane + nitrous oxide	- The TC/CC genotype is significantly associated with nausea, retching and PONV 2 to 6 h postoperatively (60% greater PONV risk compared to homozygous TT genotypes, $P=0.02$). - *KCNB2* CT/CC genotype is an independent predictor for PONV apart from the Apfel score ($P=0.031$).

PONV, postoperative nausea and vomiting.

contributing to PONV risk might be in incomplete linkage. These factors require further investigations in the future.

Presumably, genetic contributions to PONV are much more complex than previously assumed, and environmental influences, treatments, and anaesthetic regimens might modulate study results.[28] Therefore, differences in study concepts must be considered when interpreting results of SNP associations with PONV. At this point, we may rather have to take a step backwards than forwards. It is critical to investigate the connection between different study designs and the association of genetic traits to PONV as well as to the effectiveness of PONV prophylaxis.

As a limitation, this study was not powered to detect associations of the five additional SNPs investigated in the secondary study aim, making interpretation difficult. However, we found a significant association of the *DRD2* SNP with PONV that was independent in multivariate analyses. Since all SNPs investigated in our study were either associated with PONV in previous studies or additionally were selected from hypothesis-driven theorems, we relinquished a correction for multiple testing in our statistical analysis. This was a single-centre analysis and during this study, there was a personnel change in the clinic management. Thus minor changes in anaesthetic management cannot be ruled out completely. Furthermore, the PONV prophylaxis was not strictly specified in our study terms, but was individually determined by the anaesthesiologist responsible. Therefore, a lack of homogeneity in prophylaxis cannot be excluded. These points may have contributed to our inability to confirm the association of the *CHRM3* and the *KCNB2* SNPs. Our significant association of the *DRD2* SNP with PONV underscores that there is a genetic association with PONV, but it is much more complex than hypothesised.

Table 6 Comparison of different study results investigating the association of the *DRD2* SNP and PONV

Authors	Study cohort	Ethnicity	Anaesthesia	Main findings
Nakagawa *et al.* 2008[16]	$n = 1070$	Japanese	- general anaesthesia or general anaesthesia in combination with epidural anaesthesia if applicable - induction: propofol, fentanyl, and vecuronium - maintenance: Continuous infusion of propofol, in combination with a low dose of sevoflurane if necessary	- GG genotype was significant risk factor for the development of early PONV (within 6 h, $P = 0.01$).
Frey *et al.* 2015[17]	$n = 306$ (children and adults)	European	- general anaesthesia - induction: etomidate, alfentanil, and mivacurium - maintenance: sevoflurane + air	- association of GA and GG genotype with the history of PONV ($P = 0.005$). - retching and vomiting was associated with GA and GG genotype in the early (0 to 6 h) and total (0 to 24 h postoperatively) ($P = 0.022$).
Wesmiller *et al.* 2017[18]	$n = 93$ (women only)	92% Caucasian 8% African-American	- no information available	- no significant association with PONV in PACU was detected
Stamer *et al.* 2019[19]	$n = 2581$	European	- general anaesthesia - induction: propfol, fentanyl, and *cis*-atracurium - maintenance: isoflurane	- no significant association with PONV was detected up to 12 h postoperatively
Klenke *et al.* 2020[13]	$n = 465$	European	- general anaesthesia - induction: thiopentone, propofol or etomidate, fentanyl, and rocuronium or succinylcholine - maintenance: isoflurane + nitrous oxide	- no significant association with PONV was detected (2 to 6 h postoperatively)
Gloor *et al.* 2021[23]	$n = 613$	European	- anaesthesia not further specified - maintenance: continuous propofol infusion or volatile anaesthetics	- no significant association with PONV was detected within 24 and 48 h

PONV, postoperative nausea and vomiting.

Further investigations will be needed to uncover associations of *CHRM3* rs2165870, *KCNB2* rs349358 and *DRD2* rs1800497 with PONV in different ethnicities and study cohorts to throw light on potential contributions to the development of PONV and estimate their prediction accuracy. These investigations should be performed under more standardised, uniform anaesthetic conditions to allow for the examination of a potential causal relationship of anaesthesia to SNP association and to re-start the project of building a new PONV prediction score. This may also help to further elucidate the pathophysiology of PONV and the response to antiemetic prophylaxis and rescue treatment in high-risk patients. With regard to the *DRD2* SNP, it would be helpful to investigate in upcoming analyses whether the success of PONV prophylaxis with *DRD2* antagonists depends on the genotype.

Since data of genetic characteristics will presumably be more easily accessible in the future, the demand for genetically adapted therapies or anaesthetic regimens could increase. Possibly, individual genetic information could be available prior to anaesthesia as standard in the future. In a broader sense, genetically adapted therapy or prophylaxis based on this information could be beneficial to patients in anaesthesiology as it is already in other fields today. Risk profile-adapted therapy, therefore, could correspond to the demands of individual precision medicine in anaesthesiology. However, we need a) molecular studies, which characterise the above-mentioned SNPs, and b) further studies that will allow us to investigate the genetic predisposition to PONV, considering its many aspects. It is also possible that artificial intelligence and the interpretation of data from digital anaesthesia systems can help to fill these gaps.

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References

1 Gan TJ, Belani KG, Bergese S, *et al.* Fourth consensus guidelines for the management of postoperative nausea and vomiting. *Anesth Analg* 2020; **131**:411–448.
2 Watcha MF, White PF. Postoperative nausea and vomiting: its etiology, treatment, and prevention. *Anesthesiology* 1992; **77**:162–184.
3 Macario A, Weinger M, Carney S, *et al.* Which clinical anesthesia outcomes are important to avoid? The perspective of patients. *Econ Health Syst Res* 1999; **1999**:652–658.
4 Gan TJ, Sloan F, de Dear LG, *et al.* How much are patients willing to pay to avoid postoperative nausea and vomiting? *Anesth Analg* 2001; **92**:393–400.

5 Thompson HJ. The management of postoperative nausea and vomiting. *J Adv Nurs* 1999; **29**:1130–1136.
6 Schumann R, Polaner DM. Massive subcutaneous emphysema and sudden airway compromise after postoperative vomiting. *Anesth Analg* 1999; **89**:796–797.
7 Bremner WG, Kumar CM. Delayed surgical emphysema, pneumomediastinum and bilateral pneumothoraces after postoperative vomiting. *Br J Anaesth* 1993; **71**:296–297.
8 Apfel C, Läära E, Koivuranta M, *et al.* A simplified risk score for predicting postoperative nausea and vomiting. *Anesthesiology* 1999; **91**:693–700.
9 Apfel C, Korttila K, Abdalla M, *et al.* A factorial trial of six interventions for the prevention of postoperative nausea and vomiting. *N Engl J Med* 2004; **350**:2441–2451.
10 Janicki PK, Vealey R, Liu J, *et al.* Genome-wide association study using pooled DNA to identify candidate markers mediating susceptibility to postoperative nausea and vomiting. *Anesthesiology* 2011; **115**:54–64.
11 Klenke S, de Vries GJ, Schiefer L, *et al.* CHRM3 rs2165870 polymorphism is independently associated with postoperative nausea and vomiting, but combined prophylaxis is effective. *Br J Anaesth* 2018; **121**:58–65.
12 Wang J, Li Y, Zheng C, *et al.* CHRM3 rs2165870 polymorphism correlates with postoperative nausea and vomiting incidence and the efficacy of ondansetron in a Chinese Han population. *Pharmgenomics Pers Med* 2020; **13**:319–326.
13 Klenke S, de Vries GJ, Schiefer L, *et al.* Genetic contribution to PONV risk. *Anaesth Crit Care Pain Med* 2020; **39**:45–51.
14 Klenke S, Frey UH. Genetic variability in postoperative nausea and vomiting: a systematic review. *Eur J Anaesthesiol* 2020; **37**:959–968.
15 Lee SH, Kim JD, Park SA, Oh CS, Kim SH. Effects of micro-opioid receptor gene polymorphism on postoperative nausea and vomiting in patients undergoing general anesthesia with remifentanil: double blinded randomized trial. *J Korean Med Sci* 2015; **30**:651–657.
16 Nakagawa M, Kuri M, Kambara N, *et al.* Dopamine D2 receptor Taq IA polymorphism is associated with postoperative nausea and vomiting. *J Anesth* 2008; **22**:397–403.
17 Frey UH, Schnee C, Achilles M, *et al.* Postoperative nausea and vomiting: the role of the dopamine D2 receptor TaqIA polymorphism. *Eur J Anaesthesiol* 2016; **33**:84–89.
18 Wesmiller SW, Sereika SM, Bender CM, *et al.* Exploring the multifactorial nature of postoperative nausea and vomiting in women following surgery for breast cancer. *Auton Neurosci* 2017; **202**:102–107.
19 Stamer UM, Schmutz M, Wen T, *et al.* A serotonin transporter polymorphism is associated with postoperative nausea and vomiting: an observational study in two different patient cohorts. *Eur J Anaesthesiol* 2019; **36**:566–574.
20 Rueffert H, Thieme V, Wallenborn J, *et al.* Do variations in the 5-HT3A and 5-HT3B serotonin receptor genes (HTR3A and HTR3B) influence the occurrence of postoperative vomiting? *Anesth Analg* 2009; **109**:1442–1447.
21 Gudmundsson S, Singer-Berk M, Watts NA, *et al.* Variant interpretation using population databases: Lessons from gnomAD. *Hum Mutat* 2022; **43**:1012–1030.
22 Genomes Project C, Abecasis GR, Altshuler D, *et al.* A map of human genome variation from population-scale sequencing. *Nature* 2010; **467**:1061–1073.
23 Gloor Y, Czarnetzki C, Curtin F, *et al.* Genetic susceptibility toward nausea and vomiting in surgical patients. *Front Genet* 2021; **12**:816908.
24 Hayashi H, Sugino S, Hayase T, *et al.* CHRM3 Gene polymorphysm is associated with postoperative nausea and vomiting in the Japanese population. Washington: American Society of Anesthesiologists; 2012.
25 Neville MJ, Johnstone EC, Walton RT. Identification and characterization of ANKK1: a novel kinase gene closely linked to DRD2 on chromosome band 11q23.1. *Hum Mutat* 2004; **23**:540–545.
26 Gluskin BS, Mickey BJ. Genetic variation and dopamine D2 receptor availability: a systematic review and meta-analysis of human in vivo molecular imaging studies. *Transl Psychiatry* 2016; **6**:e747.
27 Smith L, Watson M, Gates S, *et al.* Meta-analysis of the association of the Taq1A polymorphism with the risk of alcohol dependency: a HuGE gene-disease association review. *Am J Epidemiol* 2008; **167**:125–138.
28 Janicki PK, Sugino S. Genetic factors associated with pharmacotherapy and background sensitivity to postoperative and chemotherapy-induced nausea and vomiting. Exp. *Brain Res* 2014; **232**:2613–2625.