

Impact of Prostaglandin F₂-Alpha and Tumor Necrosis Factor-Alpha (TNF- α) on Pain in Patients Undergoing Thermal Balloon Endometrial Ablation

Bojan Grulović¹, Martina Ribič Pucelj², Mladen Krnić³ and Višnja Kokić³

¹ General Hospital Brežice, Department of Obstetrics and Gynaecology, Brežice, Slovenia

² University of Ljubljana, University Medical Centre Ljubljana, Clinical Department of Obstetrics and Gynaecology, Ljubljana, Slovenia

³ University of Split, University Hospital Centre Split, University Department for Endocrinology, Diabetes and Metabolic Disease, Split, Croatia

ABSTRACT

The primary objective of the study was to evaluate the correlation between prostaglandin F₂-alpha and tumour necrosis factor-alpha concentration and that of pain experienced by patients undergoing thermal balloon ablation. Furthermore we evaluated the correlation between the endometrial and myometrial thicknesses and the degree of pain experienced by patients undergoing the procedure, and in addition the correlation between PGF₂-alpha, TNF-alpha and endometrial and myometrial thicknesses. Single-arm cohort study (Canadian Task force classification II-2). In University Medical Centre Ljubljana, outpatient setting, 40 perimenopausal women with dysfunctional uterine bleeding (DUB), underwent endometrial thermal balloon ablation. The thickness of the endometrium and myometrium was measured prior to surgery using a transvaginal ultrasound that provided cross-sectional images. The degree of pain was rated using the visual analogue scale (VAS) and numeric rating scale (NRS) immediately and 60 minutes after the procedure. The concentration of PGF₂-alpha and TNF-alpha in venous blood was measured prior to, at the end of and 60 minutes after the procedure. The results showed a positive correlation between the concentration of PGF₂-alpha released during endometrial ablation and the endometrial and myometrial thickness ($p > 0.01$), including the reported degree of pain ($p > 0.01$). The concentration of TNF-alpha indicates a positive correlation with the level of pain ($p > 0.05$), but is not dependent on the thicknesses of the endometrium and myometrium. Endometrial thickness correlates to the degree of pain and the prostaglandin F₂-alpha concentration. In clinical practice, performing the Gynecare ThermoChoice procedure immediately after menstruation or preoperative preparation of the endometrium using oral contraceptives enables this procedure to be performed in outpatient settings and can be considered a valuable treatment option for DUB.

Key words: thermal balloon endometrial ablation (THERMACHOICE), pain, prostaglandins, tumor necrosis factor

Introduction

Dysfunctional uterine bleeding is excessive genital tract bleeding (more than 80 mL) in the absence of any pathology^{1,2}.

Approximately 15% of clinic visits in the gynaecological practice are due to abnormal uterine bleeding^{2,3}. They occur in 20% of women between the ages of 30 and 50¹.

In 1978, Neuwirth introduced resectoscopes, which was an alternative method to hysterectomy. It was a new and minimally invasive treatment for menorrhagia – us-

ing endometrial ablation or hysteroscopic transcervical resection of the endometrium (TCRE)⁴. However, the procedure was not as well established as was expected, because it was performed under general anaesthesia, meaning that preoperative endometrial preparation with hormonal drugs was required, with 7% of cases resulting in major surgical complications. The risk of hypervolemia is described and success of the procedure depends on the experience of the surgeon^{4,5}.

Therefore, non-hysteroscopic endometrial ablation which was non-surgical became much more accepted^{6,7} endometrial ablation with circulating hot saline solution, and with different thermal balloons or bipolar electrodes^{8,9}.

Despite the established preoperative treatment by administering non-steroid anti-inflammatory drugs (NSAIDs), known as antiprostaglandins with an injection (or pill), moderate to severe pain occurs in 25% of patients¹⁰.

This requires intravenous application of analgesics or even anaesthesia as well as postoperative administration of strong analgesics¹¹. This represents a certain limitation for an exclusively ambulatory procedure.

The primary objective of the study was to evaluate the correlation between prostaglandin F₂-alpha together with tumour necrosis factor-alpha and pain in patients undergoing thermal balloon ablation.

Additional objectives involved determining the concentration of PGF₂ alpha and TNF alpha released during the procedure and analyzing correlations between the following parameters: PGF₂ alpha and TNF alpha concentrations against the degree of pain, endometrial and myometrial thicknesses and the degree of pain, PGF₂ alpha and TNF alpha concentrations against endometrial and myometrial thicknesses.

Positive correlations would imply the need for preoperative endometrial preparation using oral contraceptives or by performing a procedure immediately after menstrual bleeding. This would allow endometrial ablation, to be performed in outpatient settings as an alternative to hysterectomy.

Methods

The study was performed in the Clinical Department for Reproduction at the Department of Obstetrics and Gynaecology in Ljubljana where TC (Gynecare Thermachoice II, USA) was introduced back in 2001. It was approved by the Ethics Committee and carried out in accordance with GCP and the Declaration of Helsinki.

All women referred to our outpatient clinic over a period of one year (October 2008 – October 2009) with dysfunctional uterine bleeding were offered the ThermoChoice (TC) thermal balloon ablation. Pathological endometrial changes were excluded by performing diagnostic curettage or diagnostic hysteroscopy along with endometrial biopsy over a 6-month period prior to this study procedure. Women that had previously undergone any type of endometrial ablation were excluded from the study.

Forty women agreed to the TC procedure and gave their informed consent prior their participation in the study.

The procedure itself has been in clinical use at the mentioned department since 2001 and has been performed in accordance to the manufacturer's instructions.

Before the procedure, bimanual and speculum examination of each patient was performed, as along with a

transvaginal ultrasound examination of the uterus and adnexa. The thickness of the endometrium and myometrium was measured in cross-sectional images.

To prevent pain, non-steroid anti-inflammatory drugs (NSAIDs) are commonly used prior to the TC procedure. To discover the role of prostaglandins in the cause of pain experienced by patients in the study, we administered 100 mg of tramadol i.m instead of the NSAIDs. This does not affect the synthesis of prostaglandins. Furthermore, 5 mg of diazepam was also administered orally one hour prior to the procedure. The procedure performed was a paracervical block with 1% of Xylocain administered.

The degree of pain was measured and evaluated using the visual analogue scale (VAS) and the numeric rating scale (NRS) immediately after the procedure and 60 minutes later. PGF₂-alpha and TNF-alpha concentrations in venous blood was measured three times: at the start of the procedure, at the end (lasting 8 minutes) and 60 minutes after the procedure. The procedural times were uniform across all 40 procedures.

Determination of PGF₂-alpha and TNF-alpha concentrations were performed from frozen samples at the Clinical Institute of Clinical Chemistry and Biochemistry. The immuno-enzymatic method performed using a labelled ligand and done once for each parameter.

The correlation between endometrial and myometrial thickness, PGF₂-alpha and TNF-alpha concentrations and the degree of pain using VAS and NRS was statistically evaluated.

For the purpose of statistical analysis, a linear regression method was used. Correlation statistics (Pearson, Spearman) were performed to determine whether a statistically significant correlation existed between the different factors, e.g. the endometrial thickness and degree of pain. The paired samples t-test was used for normally distributed parameters. For parameters that were not normally distributed, nonparametric statistic methods were applied (Fridman test and Wilcoxon test).

Each patient had been informed about the aim and course of the study. Once patients decided to participate in the study, they signed an informed consent form confirming their voluntary participation.

The sample size calculation was based on a 95% confidence level estimation of the frequency of patients with moderate to severe pain (25%) after TC. With 60 TC patients introduced a year at the Department of Obstetrics and Gynaecology in Ljubljana, 40 patients included in the study meant that an estimate of this prevalence with a confidence interval of 7.0% (0.18–0.32) was possible.

Results

The baseline characteristics of the cohort studied are shown in Table 1. TC was successfully completed in all 40 women (100% of the participants).

During and 60 minutes after the procedure, the study provided different concentrations of PGF₂-alpha with a

TABLE 1
BASELINE CHARACTERISTICS OF THE PATIENTS

	Proliferative (phase 1) 14 (35%)	Secretory (phase 2) 26 (65%)	Total 40 (100%)
Mean age (years)	43.3±4.7	44.9±3.5	44.4±4.0
Uterine sounding length (<10 cm)	8.27±10.6	8.66±7.1	8.53±7.6
Endometrial thickness (mm)	6.43±3.8	9.42±5.1*	8.38±4.9
Myometrial thickness (mm)	18.29±3.8	20.9±3.5*	20.00±3.84

Data are presented as $\bar{X} \pm SD$ or N (%); *p<0.05 vs proliferative phase

statistically significant difference between the following three measurements (Fridman test): $\chi^2 = 25.56$; $df=2$; $p<0.001$. While modest changes the PGF2-alpha concentrations before and at the end of the procedure were discovered, the PGF2-alpha concentration from the third measurement was significantly lower compared to the two previous measurements, i.e. before and at the end of the procedure: ($p<0.001$ and $p<0.01$) (Paired t-test).

The TNF-alpha concentration did not change significantly for the three measurements: $\chi^2=2.52$; $df=2$; $p=0.284$ (Fridman test).

The difference in the examined parameters from the subgroups of patients based on cycle phase is showed in Table 2. In the TC patient subgroup possessing proliferative (phase 1) PGF2-alpha, there is a significant difference between the 1st and 3rd measurements ($p<0.05$). There is no significant TNF-alpha difference between any of three measurements in this patient group.

In the group possessing the secretory (phase 2) PGF2-alpha concentration, at the third measurement was significantly lower compared to both measurements – before the procedure and at the end of the procedure:

($p<0.001$ and $p<0.01$). The TNF-alpha concentration at the third measurement was significantly lower when compared to both other measurements, i.e. before and at the end of the procedure: ($p<0.05$) (Paired t-test).

Upon evaluating the PGF2-alpha and TNF-alpha concentration, and the degree of pain (according to VAS), the results showed a statistically significant positive correlation ($p<0.01$) between the PGF2-alpha concentration before the procedure and degree of pain at the end of the procedure with the whole procedure lasts for 8 minutes.

The study also discovered a statistically significant positive correlation ($p<0.01$) between the PGF2-alpha concentration measured 60 minutes after the procedure (3rd measurement) and degree of pain evaluated at the same time.

A statistically significant positive correlation ($p<0.05$) was also found between the TNF-alpha measured at 60 minutes after the procedure and degree of pain at the end of procedure. This indicates that a higher TNF-alpha concentration measured one hour after the procedure corresponds to a higher degree of pain at the end of the procedure.

TABLE 2
SUBGROUP OF TC PATIENTS WITH PROLIFERATIVE (PHASE 1) AND SECRETORY (PHASE 2)

	PGF2-alpha Difference between 2 nd and 1 st measurement	PGF2-alpha Difference between 3 rd and 1 st measurement	PGF2-alpha Difference between 3 rd and 2 nd measurement	TNF-alpha Difference between 2 nd and 1 st measurement	TNF-alpha Difference between 3 rd and 1 st measurement	TNF-alpha Difference between 3 rd and 2 nd measurement
Statistical difference (total N=40 patients)	-1.73	-3.89	-2.91	-0.64	-1.84	-1.61
p value	NS	<0.001	<0.01	NS	NS	NS
Statistical difference (phase 1, N=14 patients)	-1.55	-2.30	-1.34	-0.71	-0.03	0.66
p value	NS	<0.05	NS	NS	NS	NS
Statistical difference (phase 2, N=26 patients)	-0.93	-3.22	-2.53	-0.32	-2.12	-2.53
p value	NS	<0.001	<0.01	NS	<0.05	<0.05

Differences between three measurements of PGF2-alpha in TNF-alpha concentrations (NS=not statistically significant)

In evaluating the correlation between the endometrial/myometrial thickness and the degree of pain, positive correlations were also calculated (Figures 1 and 2).

Figure 1 shows that a thicker endometrium is associated with higher levels of pain during and after the procedure ($p < 0.01$), whereas Figure 2 shows the same for a thicker myometrium.

A positive correlation ($p < 0.01$) between the PgF2-alpha concentration before the procedure against endometrial and myometrial thicknesses was also calculated.

The PGF2-alpha concentration before the procedure correlates to the thicknesses of the endometrium and myometrium and the degree of pain.

A correlation between the quantity of TNF-alpha and endometrium and myometrium thicknesses was not observed.

Discussion

The purpose of our study was to determine the correlation between the prostaglandin F2-alpha (PGF2-alpha) concentration and tumour necrosis factor-alpha (TNF-alpha), against the degree of pain together with endometrial and myometrial thicknesses in patients undergoing TC thermal balloon ablation.

For many years prostaglandins have been recognized as key molecules in endometrial physiology and in endometrial glands and menstruation. More recently, a role for COX enzymes and prostaglandins has been ascertained in reproductive tract pathology, including dysmenorrhea, endometriosis and menorrhagia¹².

The cause of pain is not completely known but it is mainly attributed to prostaglandins, particularly PGF2-alpha¹³ and is commonly used in preoperative treatment in which upon the administering of antiprostaglandin agents – NSAIDs, pain is significantly reduced.

On the other hand, Laberge et al. 2003 in a prospective double-arm study, comparing intraoperative and postoperative pain in relation with two modes of non-hysteroscopic ablation, showed that frequency and the degree

of pain depend on the duration of the procedure (NovaSure, Hologic, USA 2 min., TC 8 min.) and not on the amount of prostaglandins released. The shorter procedure results in a lower degree of pain¹⁴. The question which remains is why the degree and intensity of pain greatly differs in the same group of patients undergoing the same (TC) procedure.

In our study, we found that the endometrial and myometrial thicknesses show a positive correlation when compared to the degree of both intraoperative and postoperative pain (Figures 1 and 2).

A statistically significant difference among the three PGF2-alpha measurements was found. The third PGF2-alpha concentration was in statistical terms significantly lower (Table 2). This finding indicates that during endometrial ablation with TC, PGF2-alpha is released and shows a positive correlation to the degree of pain recorded on the VAS and NRS scales.

Higher concentrations of PGF2-alpha before the procedure are released from the thicker endometrium and myometrium and correspond to a higher degree of pain during and after the procedure (2nd measurement). This is confirmed by our results, because none of the patients with an endometrium thicker <5 mm experienced pain >5 according to VAS (Table 3) (Fisher exact test $p < 0.001$).

Furthermore, the degree of pain was significantly lower in patients with an endometrial thickness <5 mm.

Significantly lower concentrations of PGF2-alpha one hour after the procedure corresponded to a lower degree of pain at that same time.

Tumour necrosis factor alpha (TNF-alpha), one of the tumour markers for endometriosis, is released from the endometrium during the secretory phase and causes hemorrhagic cell necrosis^{15,16}.

TNF-alpha was released during and after the procedure but not to the extent observed with PGF2-alpha, and did not show any statistically significant differences in concentrations (Table 2). These concentrations remained stable throughout three measurements.

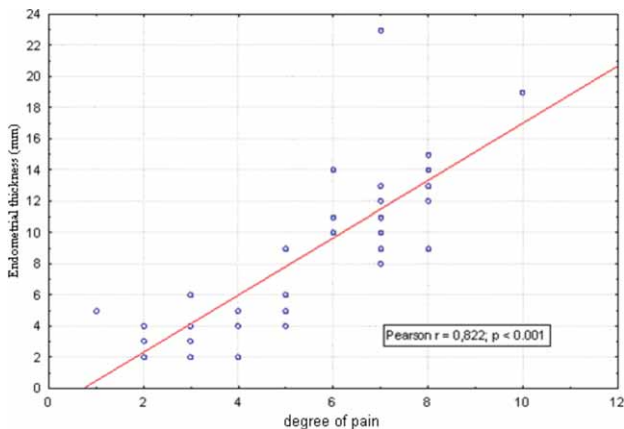


Fig. 1. Correlation between endometrial thickness (mm) and the degree of pain (VAS score) during the procedure.

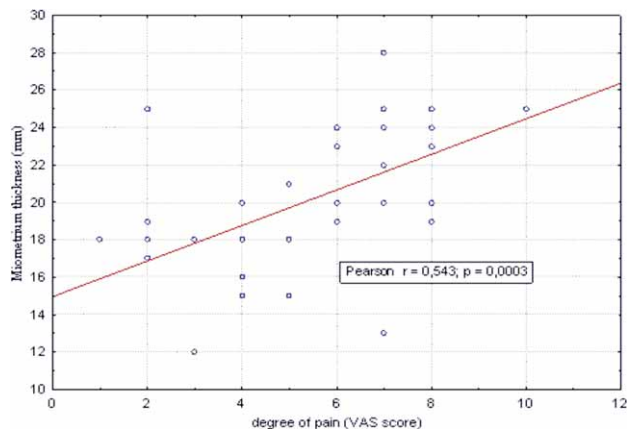


Fig. 2. Correlation between myometrial thickness (mm) and the degree of pain (VAS score) during the procedure.

TABLE 3
CORRELATION OF ENDOMETRIAL THICKNESS AND PAIN SCORE

	Endometrial thickness <5 mm	Endometrial thickness >5 mm
N°of patients	15	25
N°of patients VAS I score>5	0**	25
N°of patients VAS II score>5	0**	25
Average pain score VAS I	3.13±1.18	6.60±1.52##
Average pain score VAS II	2.00±1.64	3.56±1.96#

* Data are presented as N or $\bar{X}\pm SD$; ** Fisher exact test $p<0.001$; # Student's t-test $p<0.05$, ## Student's t-test $p<0.01$.

Higher TNF-alpha concentrations one hour after the procedure showed a correlation with a higher degree of pain during and after the procedure. However, the TNF-alpha concentration showed no correlation with endometrial and myometrial thicknesses.

A possible objection to the study could be that the PGF2-alpha samples were obtained from venous blood. This is particularly so if given that most of the prostaglandins metabolized during the first passage through the lungs. However, the purpose of the study was to find only changes in PGF2-alpha concentrations and not the absolute values released, which was less invasive and aggravating for patients. Further studies will be necessary to confirm these findings.

Ultrasound (US) measurements of the cross-sectional images were performed by two authors from the US, both highly skilled and experienced. In addition, 40 consecutive patients who fulfilled inclusion criteria were recruited in the study during over a one-year period.

In the treatment of menorrhagia TC represents an efficient alternative to the significantly more invasive hysterectomy^{17,18} and can be performed in 87% of patients in outpatient settings¹⁹.

Finding from previous studies showed that administration of an oral contraceptive pill (OC) in the proliferative phase of the cycle maintains a very thin, flat endometrium²⁰.

Therefore, in performing the thermal endometrial ablation procedure immediately after the menstruation, or if preparation of endometrium is done preoperatively with oral contraceptives (if not contraindicated), it would allow the procedure to be performed in outpatient settings without the need for anaesthesia.

This assumption should be further tested and validated in a future well thought out study.

Conclusions

The intraoperative and postoperative degree of pain in women undergoing TC endometrial ablation is in direct correlation with the released PGF2-alpha, and which subsequently shows a positive correlation with endometrial and myometrial thicknesses.

Acknowledgements

This study was partially sponsored by Johnson & Johnson S.E., Slovenia. The authors are grateful to all the staff and women who participated in this research study. Special thanks to Prof. Dr. Joško Osredkar, Head of Institute of Clinical Chemistry and Biochemistry. The authors declare there are no conflicts of interest, financial or otherwise. The authors alone are responsible for the content and the writing of the paper.

REFERENCES

- RIBIČ-PUCELJ M, Endoscopic Surgery in Gynaecology (Didakta, Ljubljana, 2007). — 2. FARELL E, Aust Fam Physician, 33 (2004) 906. — 3. BONGERS MY, MOL BW, BROLMANN HA, Maturitas, 47 (2004) 159. DOI: 10.1016/j.maturitas.2003.08.002. — 4. VILOS GA, Obstet Gynecol Clin North Am, 31 (2004) 687. DOI: 10.1016/j.ogc.2004.05.009. — 5. BOURDES P, BONGERS MY, MOL BW, Fertile Sterile, 82 (2004) 160. DOI: 10.1016/j.fertnstert.2003.12.025. — 6. WASZAK SJ, J Reprod Med, 49 (2004) 769. — 7. CLARK TJ, GUPTA JK, Fertile Sterile, 82 (2004) 1395. DOI: 10.1016/j.fertnstert.2004.04.042. — 8. SHAAMASH AH, SAYED EH, J Obstet Gynecol Res, 30 (2004) 210. DOI: 10.1111/j.1447-0756.2004.00189.x. — 9. MUNRO MG, J Am Assoc Gynecol Laparosc, 11 (2004) 8. DOI: 10.1016/S1074-3804(05)60004-7. — 10. MARSH F, THEWLIS J, DUFFY S, Fertile Sterile, 83 (2005) 715. DOI: 10.1016/j.fertnstert.2004.08.030. — 11. HASSAN L, GANNON MJ, Best Pract Res Clin & Obstet Gynecol, 19 (2005) 681. DOI: 10.1016/j.bpobgyn.2005.06.008. — 12. SMITH SK, Baillieres Clin Obstet Gynecol, 3 (1989) 249. DOI: 10.1016/S0950-3552(89)80021-5. — 13. STEFANCZYK-KRZYMOWSKA S, WASOWSKA B, GILUN P, Reprod Domest Anim, 47 (2012) 98. DOI: 10.1111/j.1439-0531.2011.01807.x. — 14. LABERGE PY, SABBAH R, FORTIN C, J Am Assoc Gynecol Laparosc, 10 (2003) 223. DOI: 10.1016/S1074-3804(03)80110-X. — 15. HAIDER S, KNOFLER M, Placenta, 30 (2009) 111. DOI: 10.1016/j.placenta.2008.10.012. — 16. JUKIĆ T, ABIDOV M, IHAN A, Coll Antrop, 35 (2011) 1219. — 17. STRINIĆ T, VULIĆ M, BUKOVIĆ D, MASKOVIĆ J, HAUPTMAN D, JELINCIĆ Z, Coll Antropol, 28 (2004) 793. — 18. ELGARIB AE, NOOH A, J Obstet Gynecol, 26 (2006) 669. DOI: 10.1080/01443610600913882. — 19. MARSH F, THEWLIS J, DUFFY S, Fertile Steril, 87 (2007) 642. DOI: 10.1016/j.fertnstert.2006.07.1502. — 20. GROW DR, IROMLOO K, Fertile Sterile, 85 (2006) 204. DOI: 10.1016/j.fertnstert.2005.06.044.

B. Grulović

General Hospital Brežice, Department of Obstetrics and Gynaecology, Prešernova cesta 17, 8250 Brežice, Slovenia
e-mail: bojan.grulović@zg.t-com.hr

UTJECAJ PROSTAGLANDINA F2-ALFA FAKTORA TUMORSKE NEKROZE-ALFA (TNF- α) NA BOL U PACIJENATA PODVRGNUTIH TERMALNOJ ENDOMETRIJSKOJ BALON ABLACIJI

S A Ž E T A K

Osnovni cilj istraživanja bio je procijeniti korelaciju između prostaglandina F2-alfa i nekroze tumora koncentracije faktor-alfa te boli koju osjećaju bolesnici podvrgnuti toplinskoj balon ablaciji. Nadalje, procijenili smo korelaciju između endometrijske i miometrijske debljine i stupnja boli koju doživljavaju pacijenti koji prolaze postupak, a osim toga i korelaciju između PGF2-alfa, TNF-alfa i endometrijske i miometrijske debljine. Provedeno je jednostruko kohortno istraživanje (Canadian Task force classification II-2) u sveučilišnom medicinskom centru u Ljubljani, u dnevnim bolnicama. 40 žena pred menopauzu s disfunkcionalnim krvarenjem iz maternice (DUB), prošle su endometrijalnu toplinsku balon ablaciju. Endometrijska i miometrijska debljina mjerena je prije operacije koristeći transvaginalni ultrazvuk koji pruža presjek slike. Stupanj boli ocijenio se pomoću vizualne analogne skale (VAS) i brojčane skale ocjena (NRS) neposredno nakon i 60 minuta nakon zahvata. Koncentracija PGF2-alfa i TNF-alfa u venskoj krvi mjerene su prije, na kraju, i 60 minuta nakon postupka. Rezultati su pokazali pozitivnu korelaciju između koncentracije PGF2-alfa tijekom ablacije endometrija i endometrijske i miometrijske debljine ($p < 0,01$), uključujući i izmjeren stupanj boli ($p < 0,01$). Koncentracija TNF-alfa pokazuje pozitivnu korelaciju s razinom boli ($p < 0,05$), ali nije ovisna o endometrijskoj i miometrijskoj debljini. Endometrija debljina korelira sa stupnjem boli i koncentracijom prostaglandina F2-alfa. U kliničkoj praksi, izvođenje postupka Gynecare ThermaChoice odmah nakon menstruacije ili predoperativne pripreme endometrija korištenjem oralnih kontraceptiva omogućuje da se ovaj postupak obavlja u ambulantnim uvjetima i može se smatrati vrijednim tretmanom za DUB.