# Spectroscopy Analysis of Uterine Leiomyomas Before and After Treatment with GnRH-a

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MR spectroscopy uses the same hardware as magnetic resonance imaging but provides information at the biochemical level. It has been used to measure proton-containing compounds, such as amino acids, fatty acids, organic acids, sugars and other metabolic active compounds in tissue specimens. Before and after GnRH-a treatment we measured lipid, lactate, creatine and choline concentrations in two patients with uterine leiomyomas using single-voxel proton MR spectroscopy. Subcutaneous goserelin acetate injection was performed at least two times every 4 weeks before surgery. Spectroscopy was performed 3 weeks later last goserelin acetate injection. The MR spectroscopy results of leiomyomas before and after GnRH-a treatment were compared. Before GnRH-a treatment, spectral analysis of each patients showed only choline signals. After GnRH-a treatment, spectrum of both patients showed lipid peaks but the other signals were not observed. The presence of lipid signals and absence of other signals suggest a diffuse metabolic defect in association with GnRHa treatment, possibly consistent with decrease in myoflaments, and increase in apopitosis and necrosis.
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**Key Words:** Uterine leiomyoma, Gonadotropin-releasing hormone, Magnetic resonance spectroscopy

In vivo proton magnetic resonance spectroscopy (MRS) addresses metabolic pathways in different tissues and it provides information about the contents of organic compounds in living tissues and lead to a better understanding of the biochemical pathways found within a lesion.[1-3] Despite the increasing knowledge about various brain diseases on MR spectroscopy, the role of this modality in distinguishing different types of pelvic diseases has been limited because of the technique reasons. GnRH agonists (GnRH-a), which induce medical castration, are now being used to treat uterine leiomyomas.[4,5] Most uterine leiomyomas shrink as a result of administration of GnRH-a.[4,5] Although only isolated report of MR spectroscopy of uterine leiomyoma has been published[6] the MR spectroscopy of uterine leiomyomas before and after GnRH-a treatment has not been studied. We herein report the findings of semiquantitative proton MR spectroscopic imaging of two patients with uterine leiomyomas. The aim of this study to compare spectroscopy features of leiomyoma cells from the same uterine myoma nodule before and after GnRH-a treatment. To our knowledge, this is the first report to examine the levels of choline, creatine, lipid and lactate in uterine leiomyoma before and after GnRH-a treatment.

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## Cases

Two patients with symptomatic uterine myoma included in this study. Their symptoms were abnormal uterine bleeding, pelvic pain and palpable abdominal mass. Each of them had undergone MRS immediately after gynecologic and ultrasonographic examination. The gynecologic examination of two patients revealed a bigger uterus than the normal size. Globally enlarged uterus (approximately 18 cm x 18 cm) was seen at transvaginal ultrasonography (TV-US) of patient 1. The gynecologic examination of the other patient showed a bigger and irregular uterus than the normal size and shape. TV-US revealed multiple leiomyomas that one placed in the cervix and remaining leiomyomas located in the corpus and fundus uterus. The diameter of the uterine fibroids ranged from 3 to 8 cm Goserelin acetate (Zoladex; Astra Zeneca PLC-UK) was administered at subcutaneously every 4 weeks, for a period of 8 weeks. The MRS was performed 3 weeks later the last goserelin acetate injection. After MRS each of the patients has undergone total abdominal hysterectomy. The metabolites level of uterine leiomyoma cells were calculated by spectroscopy. MRS (Philips, Best, The Netherlands) was performed by the point resolved spectroscopy technique with a long TE (TR= 1500 msec, TE= 136 msec). The voxel volume size was 2x2x2 $cm^3$ The region of interest for MRS measurement was placed at the centre of the uterine leiomyomas to avoid a contamination signal from peripheral tissues. Metabolite peak areas were calculated using a simplex routine, assuming gaussian line shapes. The semiquantified evaluation of creatine, choline, lactate and lipid were conducted by the specialist of radiology and MRS. The metabolites level were classified into three classes, in comparison with the noise level by visual estimation: that is, twofold higher than the average noise level (++), higher than the average noise level but lower than a twofold

higher noise level (+), and the same as the average noise level (-). The classification of metabolite concentrations in this study were performed according to the technique described by Okada et al.[3] Table 1 describes the semiquantitative evaluation of the metabolite peak areas in two cases. Before GnRH-a treatment, spectral analysis of each patients showed only choline signals (Figure 1a and 2a). After GnRH-a treatment, spectral analysis of both patients showed lipid peaks (Figure 1b and 2b) but the other signals were not observed

*Table 1. Semi-quantitative evaluation of the metabolite levels in uterine leiomyomas before and after GnRH-a treatment.*

| Patients | Age | Metabolites | Before GnRH-a | After GnRH-a |
|---|---|---|---|---|
| 1 | 26 | Choline | ++ | - |
| | | Creatine | - | - |
| | | Lactate | - | - |
| | | Lipid | - | ++ |
| 2 | 30 | Choline | ++ | - |
| | | Creatine | - | - |
| | | Lactate | - | - |
| | | Lipid | - | ++ |

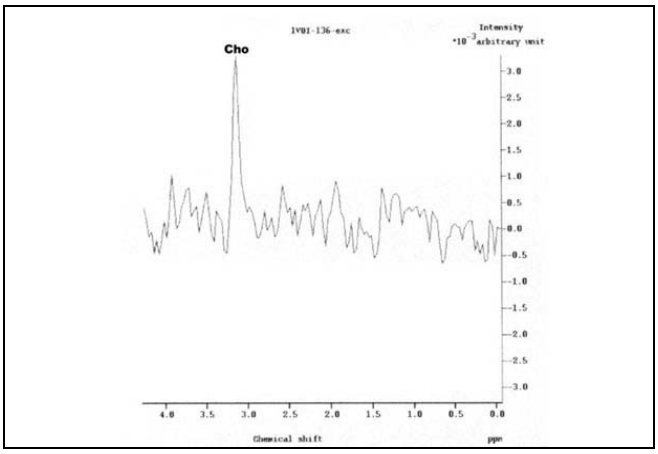


Figure 1a. Proton MR spectroscopy in case of uterine leiomyoma before GnRH-a treatment (TR= 1500 msec, TE= 136 msec). The high Cho peak was detected.

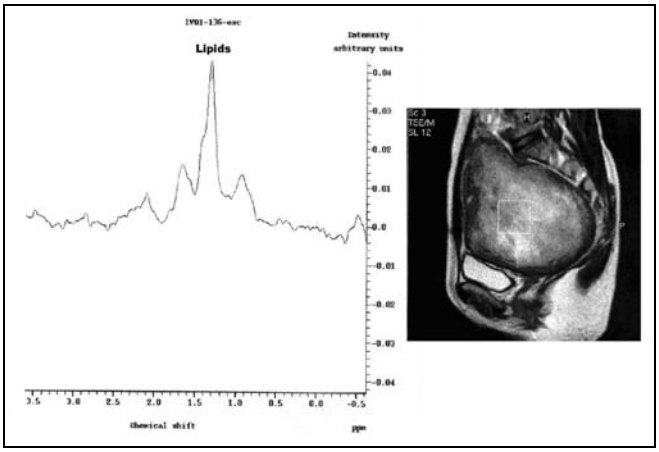


Figure 1b. Proton MR spectroscopy in case of uterine leiomyoma after GnRH-a treatment (TR= 1500 msec, TE= 136 msec). The high lipid signal was detected, but the signals of Cho, Cr and lactate were not observed.

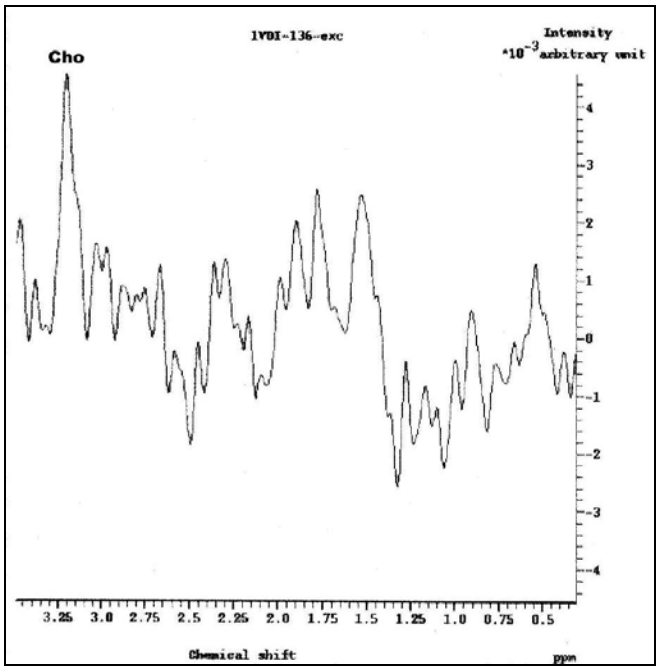


Figure 2a. Proton MR spectroscopy in case of uterine leiomyoma before GnRH-a treatment (TR= 1500 msec, TE= 136 msec). The high choline peak was detected.

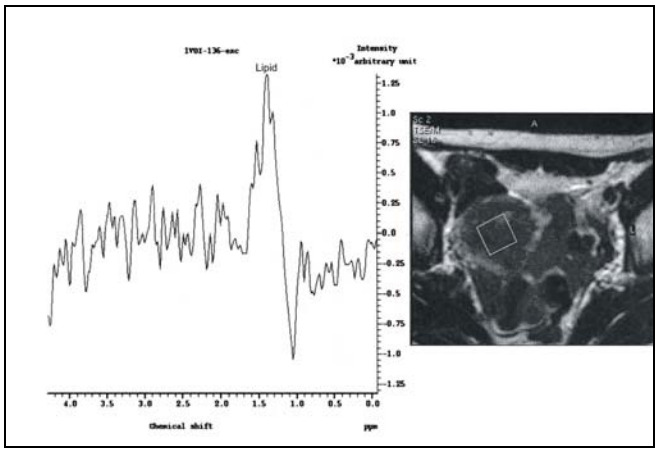


Figure 2b. Proton MR spectroscopy after GnRH-a treatment (TR= 1500 msec, TE= 136 msec). The only high lipid signal was detected.

## Conclusion

MR imaging has proved to have a greater potential in showing the size, number, and location of leiomyomas and the presence of the degeneration.[7] Also, it can be used to document the regression of leiomyomas after treatment with GnRH-a.[7,8] MRS is a procedure in which nuclear MR signals are obtained from nuclei other than water. It uses the same hardware as MR imagine but provides information at the biochemical level. The spectrum allows absolute quantification of metabolite concentration and provides structural information regarding various molecular species.[9] NAA, Cho, Cr, lactate and lipid were found as an important metabolites in the spectra. NAA is generally recognized as a marker of functional neurons and their appendages.[10]

Choline is a cell membrane and myelin marker and it plays an important role in the structure and biochemical activity of cell membranes.[11-13] An increase in choline signal represents increased membrane synthesis and cell turnover rates. A recent study Okada et al.[3] demonstrated creatine, choline and lactate signals in the spectral analysis of three patients with uterine leiomyomas. However, none of them showed lipid signals. In the present study, spectroscopy analysis of two patients before GnRH-a treatment displayed only choline signals. Since choline is thought to be a cell membrane marker, the increased choline signals may be the result of alteration in membrane metabolism of leiomyoma cells. Another possible explanation may be that, its elevation could reflect increased membrane turnover or changes in cellular density or composition of leiomyoma cells.

Lactate signal demonstrates a mismatch between glycolysis and oxygen supply.[14,15] An increase in lactate signal suggests a dependence on the anaerobic metabolism. Cr is considered to be an indicator of brain energy metabolism.[16] Spectral analysis of two patients before and after GnRH-a treatment did not show creatine and lactate signals.

MR visible lipids in normal tissues are bound to macromolecules in membranes. Therefore, lipids are physiologically not detectable in the spectrum of healthy human. Due to some pathologic disruption of such structures, increased lipid turnover leads to more mobile lipids, yielding lipid peaks at 0.9 and 1.3 ppm.[17] Rapid cellular turnover in malignant tissue usually leads to a considerable amount of cellular death, resulting in areas of necrosis which appears in MRS as a lipid signal.[17] In tumors of the brain and uterine cervix, an elevated lipid peak, as demonstrated by MRS, is thought to be a possible indicator of malignancy.[2,18] Interestingly, spectroscopy obtained from two patients after GnRH-a treatment showed only lipid signals at 1.3 ppm, but the other signals were not observed in the spectrum. This findings may be the result of GnRH-a treatment. Ito et al.[19] showed that cellular atrophy due to a decrease in myofilaments plays a major role in the myoma shrinkage resulting from GnRH-a treatment. Because hypoestrogenic environment due to GnRH-a may lead to a subsequent decrease in the amount of blood supply to the leiomyoma cells.[20] Kalir et al.[21] reported that myomas from patients treated with GnRH-a demonstrated more hyalinization, and larger collagen fibrils than those of age-matched control patients. In addition to above mentioned studies, several investigations have shown more necrosis and apoptosis in uterine leiomyoma tissue as a result of GnRH-a treatment.[22,23] A recent study has shown that the amount of lipids detected by MRS correlates well with the degree of necrosis[17] seen on histology. Elevated levels of lipid reflect lipid release from phospholipids membrane as a result of the break up of cell membranes. For that reason, the presence of lipid signal after GnRH-a treatment may be the result of necrosis or pathologic disruption of leiomyoma cell structures.

In both cases reported herein, proton MR spectroscopic imaging showed intense lipid signals. The current study indicates that a diffuse, probably global, metabolic abnormality occurs in associated with GnRH-a treatment. The most pronounced change was an increase in lipid signal after GnRH-a treatment. A rise in lipid can have multiple causes, including increased apoptosis and necrosis, decreased blood supply and myofilaments density and more hyalinization. However, this explanation must be regarded as speculative because histopathologic studies of leiomyomas after GnRHa treatment are scarce and no evidence of degeneration and necrosis were found in lesions of two patients. Future studies of larger numbers of patients using other techniques are required to confirm or refute our findings. Finally, in vivo proton MRS may be helpful better understanding of the metabolic and ultrastructural changes induced in uterine leiomyomas by GnRH-a.

## References


1. Burtscher IM, Holtas S. Proton MR spectroscopy in clinical routine. J Magn Reson Imaging 2001; 13:560-7.
2. Gotsis ED, Fountas K, Kapsalaki E, Toulas P, Peristeris G, Papadakis N. In vivo proton MR spectroscopy: the diagnostic possibilities of lipid resonances in brain tumors. Anticancer Res 1996; 16:1565-7.
3. Okada T, Harada M, Matsuzaki K, Nishitani H, Aono T. Evaluation of female intrapelvic tumors by clinical proton MR spectroscopy. J Magn Reson Imaging 2001; 13:912-7.
4. Adamson GD. Treatment of uterine fibroids: current findings with gonadotropin-releasing hormone agonists. Am J Obstet Gynecol 1992; 166:746-51.
5. Friedman AJ, Lobel SM, Rein MS, Barbieri RL. Efficacy and safety considerations in women with uterine leiomyomas treated with gonadotropin-releasing hormone agonists: the estrogen threshold hypothesis. Am J Obstet Gynecol. 1990; 163:1114-9.
6. Celik O, Sarac K, Hascalik S, Alkan A, Mizrak B, Yologlu S. Magnetic Resonance Spectroscopy Features of Uterine Leiomyomas. Gynecol Obstet Invest. 2004; 29 [Epub ahead of print]
7. Hricak H, Tscholakoff D, Heinrichs L, et al. Uterine leiomyomas: correlation of MR, histopathologic findings, and symptoms. Radiology 1986; 158:385-91.
8. Persaud V, Arjoon PD. Uterine leiomyoma. Incidence of degenerative change and a correlation of associated symptoms. Obstet Gynecol 1970; 35:432-6.
9. Van Zijl PC, Barker PB. Magnetic resonance spectroscopy and spectroscopic imaging for the study of brain metabolism. Ann N Y Acad Sci 1997; 820:75-96.
10. Barker PB, Glickson JD, Bryan RN. In vivo magnetic resonance spectroscopy of human brain tumors. Top Magn Reson Imaging 1993; 5:32-45.

11. Tien RD, Lai PH, Smith JS, Lazeyras F. Single-voxel proton brain spectroscopy exam (PROBE/SV) in patients with primary brain tumors. AJR Am J Roentgenol. 1996; 167:201-9.

12. Kugel H, Heindel W, Ernestus RI, Bunke J, du Mesnil R, Friedmann G. Human brain tumors: spectral patterns detected with localized H-1 MR spectroscopy. Radiology 1992; 183:701-9.

13. Go KG, Kamman RL, Mooyaart EL, et al. Localised proton spectroscopy and spectroscopic imaging in cerebral gliomas, with comparison to positron emission tomography. Neuroradiology. 1995; 37:198-206.

14. Alger JR, Frank JA, Bizzi A, et al. Metabolism of human gliomas: assessment with H-1 MR spectroscopy and F-18 fluorodeoxyglucose PET. Radiology 1990; 177:633-41.

15. Lanfermann H, Kugel H, Heindel W, Herholz K, Heiss WD, Lackner K. Metabolic changes in acute and subacute cerebral infarctions: findings at proton MR spectroscopic imaging. Radiology 1995; 196:203-10.

16. Sutton LN, Wang Z, Gusnard D, et al. Proton magnetic resonance spectroscopy of pediatric brain tumors. Neurosurgery 1992; 31:195-202.

17. Kuesel AC, Briere KM, Halliday WC, Sutherland GR, Donnelly SM, Smith IC. Mobile lipid accumulation in necrotic tissue of high grade astrocytomas. Anticancer Res 1996; 16:1485-9.

18. Lee JH, Cho KS, Kim YM, et al. Localized in vivo 1H nuclear MR spectroscopy for evaluation of human uterine cervical carcinoma. AJR Am J Roentgenol 1998; 170:1279-82.

19. Ito F, Kawamura N, Ichimura T, Tsujimura A, Ishiko O, Ogita S. Ultrastructural comparison of uterine leiomyoma cells from the same myoma nodule before and after gonadotropin-releasing hormone agonist treatment. Fertil Steril 2001; 75:125-30.

20. Spong CY, Sinow R, Renslo R, Cabus E, Rutgers J, Kletzky OA. Induced hypoestrogenism increases the arterial resistance index of leiomyomata without affecting uterine or carotid arteries. J Assist Reprod Genet 1995; 12:338-41.

21. Kalir T, Goldstein M, Dottino P, et al. Morphometric and electron-microscopic analyses of the effects of gonadotropin-releasing hormone agonists on uterine leiomyomas. Arch Pathol Lab Med 1998; 122:442-6.

22. Colgan TJ, Pendergast S, LeBlanc M. The histopathology of uterine leiomyomas following treatment with gonadotropin-releasing hormone analogues. Hum Pathol 1993; 24:1073-7.

23. Mizutani T, Sugihara A, Nakamuro K, Terada N. Suppression of cell proliferation and induction of apoptosis in uterine leiomyoma by gonadotropin-releasing hormone agonist (leuprolide acetate). J Clin Endocrinol Metab 1998; 83:1253-5.