

The decline in serum choline concentration in humans during and after surgery is associated with the elevation of cortisol, adrenocorticotrophic hormone, prolactin and β -endorphin concentrations

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Abstract

Serum choline concentrations decrease during and after surgery. We undertook this study to determine whether the decrease of choline is associated with an increase in stress hormones. In 16 patients undergoing abdominal surgery with general anesthesia, circulating choline, cortisol, prolactin, adrenocorticotrophic hormone (ACTH) and β -endorphin levels were measured before, during and after surgery. Choline levels decreased by 41% ($P < 0.01$) during surgery, remained 15–38% decreased for 48 h, and returned to preoperative values 72 h after surgery. The decrease in serum choline was associated and inversely correlated with the increase in serum cortisol ($P < 0.001$; $r = -0.642$), prolactin ($P < 0.001$; $r = -0.756$), β -endorphin ($P < 0.001$; $r = -0.726$) and ACTH ($P < 0.01$; $r = -0.458$). In conclusion, we found that abdominal surgery induces a decline in serum choline associated with an increase in circulating cortisol, prolactin, ACTH and β -endorphin. © 2002 Published by Elsevier Science Ireland Ltd.

Keywords: Choline; Surgery; Stress; Cortisol; Prolactin; Adrenocorticotrophic hormone; β -Endorphin

Choline is a biocation having roles in several vital physiological processes. It is the precursor of phosphatidylcholine and sphingomyelin, two phospholipids that are major components of biological membranes and precursors for intracellular messengers such as diacylglycerol or ceramide [2,18]. Choline is also the precursor of signaling lipids, platelet-activating factor and sphingosylphosphorylcholine, and of a neurotransmitter, acetylcholine [2,12,18]. Furthermore, choline is enzymatically oxidized to betaine, an important methyl donor. Circulating choline affects acetylcholine release [3,8,14] and cholinergic neurotransmission [13], as well as phospholipid synthesis and levels [11].

Recently, we demonstrated that serum choline concentrations decrease in humans after surgery, childbirth and traumatic head injury [16]. In light of this finding and of the vital

role of choline in cell membrane synthesis and cholinergic neuron function, in the present study, we measured changes in serum choline levels during and after surgery in a larger group of patients. We also sought to determine whether the decrease in serum choline is related to intra- and postoperative elevations in the stress hormones cortisol, adrenocorticotrophic hormone (ACTH), β -endorphin and prolactin [4–7,9,10].

Following institutional approval (Uludag University Ethics Committee) and written informed consent, 16 patients scheduled for elective major abdominal or gynecological surgery with general anesthesia were included in the analysis. Anesthesia was induced with intravenous sodium thiopental (5–10 mg/kg) administration and maintained with either halothane/nitrous oxide or isoflurane/nitrous oxide. All surgeries were performed at the Uludag University Teaching and Research Hospital (Bursa, Turkey).

Venous blood samples were collected in glass Vacutainer

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tubes (for serum choline and cortisol) and in plastic Vacutainer tubes containing ethylene-diaminetetraacetic acid (for plasma ACTH and β -endorphin) at the following time intervals: before surgery (within 2 h of surgery); at the surgical onset (5–10 min after incision); intraoperatively (45–90 min after incision); upon admission to the recovery room; and at 1, 6, 12, 24, 36, 48 and 72 h postoperatively. Blood samples were immediately placed on ice and centrifuged within 30 min at 4 °C at 1500 \times g for 10 min. Serum and plasma samples were stored at –20 °C until analysis. Serum choline was measured radioenzymatically [17] as described previously [16]. Serum cortisol and prolactin and plasma ACTH were measured by a solid phase chemiluminescent enzymeimmunoassay system using a commercially available kit (Bio DPC, Los Angeles, CA). Plasma β -endorphin was extracted and measured by radioimmunoassay (Incstar, USA).

Preoperatively, the mean serum choline level was $11.3 \pm 0.9 \mu\text{M}$ (Table 1). During and immediately after surgery, serum choline levels significantly decreased by 41 and 22%, respectively. Postoperatively, serum choline levels remained decreased by 15–38% for 48 h (Table 1). Serum choline levels returned the preoperative value 72 h after surgery (Table 1).

Before surgery, mean values for stress hormones were: serum cortisol, $12.9 \pm 1.8 \mu\text{g/dl}$ ($n = 16$); serum prolactin, $9.6 \pm 1.9 \text{ ng/ml}$ ($n = 16$); plasma ACTH, $21.4 \pm 3.6 \text{ pg/ml}$ ($n = 16$); and plasma β -endorphin, $14.6 \pm 1.7 \text{ pg/ml}$ ($n = 10$). As seen in Table 1, significant elevations were observed in the concentrations of these hormones at several time points during and after surgery.

Fig. 1 shows the relationship of serum cortisol, serum prolactin, plasma ACTH and plasma β -endorphin concentrations to serum choline concentrations. Serum choline levels

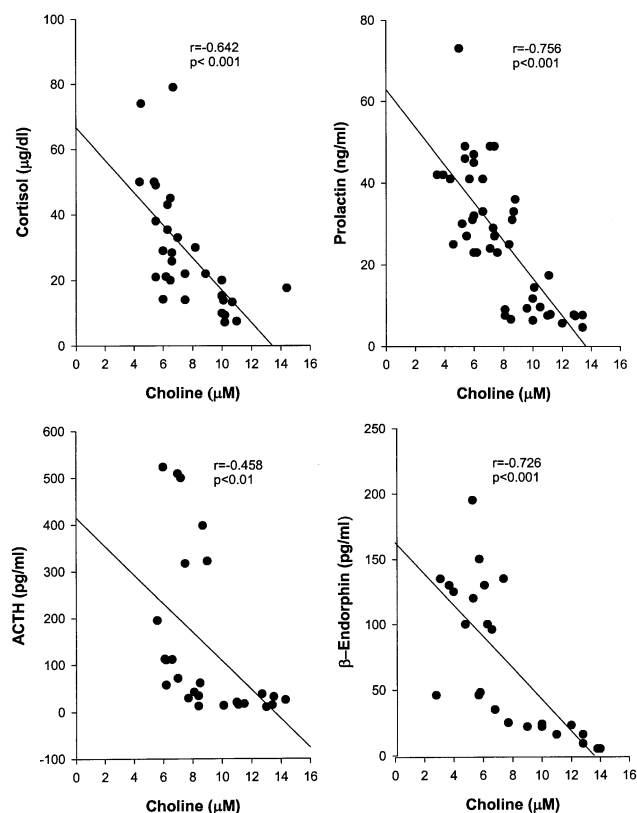


Fig. 1. The relationship of serum choline concentrations to serum cortisol, serum prolactin, plasma ACTH and plasma β -endorphin concentrations. Serum choline concentrations in venous blood samples obtained before, during, and following surgery were plotted against the concentrations of cortisol, prolactin and β -endorphin collected at the same time intervals. For choline and ACTH, serum choline concentrations in blood samples were plotted against the plasma ACTH concentrations obtained before surgery, and at 1 and 6 h after surgery.

Table 1

Changes in choline, cortisol, prolactin, ACTH and β -endorphin concentrations intra- and postoperatively^a

Periods	Choline (μM)	Cortisol ($\mu\text{g/dl}$)	Prolactin (ng/ml)	ACTH (pg/ml)	β -Endorphin (pg/ml)
Preoperative	11.3 ± 0.9	12.9 ± 1.8	9.6 ± 1.9	21.4 ± 3.6	14.6 ± 1.7
Incision	10.7 ± 0.8	23.2 ± 1.0^b	9.0 ± 0.8	ND	32.3 ± 4.6^c
Intraoperative	6.9 ± 0.7^d	33.2 ± 1.0^b	30.3 ± 4.8^d	ND	59.1 ± 5.1^d
Recovery room admission	8.8 ± 0.7^d	32.0 ± 3.7^b	40.7 ± 5.7^d	ND	43.4 ± 3.6^d
Postoperative					
1 h	8.8 ± 1.0^d	36.1 ± 5.0^b	51.1 ± 8.0^d	245.6 ± 70.0^c	ND
6 h	8.0 ± 1.2^d	31.8 ± 2.9^b	31.1 ± 3.7^d	106.0 ± 41.0^c	ND
12 h	7.0 ± 0.9^d	19.0 ± 1.9^c	25.5 ± 3.1^d	25.5 ± 6.0	ND
24 h	7.8 ± 0.9^d	17.5 ± 1.6^c	27.2 ± 3.5^d	14.5 ± 1.5	9.2 ± 0.7
36 h	8.1 ± 0.9^d	15.1 ± 2.0	ND	15.4 ± 2.5	ND
48 h	9.6 ± 0.8^c	9.7 ± 2.5	24.8 ± 3.5^d	16.8 ± 3.5	9.5 ± 0.7
72 h	11.1 ± 1.1	15.1 ± 1.1	ND	ND	ND

^a Venous blood sample were obtained 0.5–1 h before anesthesia ('preoperative'), 5–10 min after the incision ('incision'), 45–90 min into surgery ('intraoperative'), within 5 min of transferring patients to the recovery room ('recovery room admission'), and 1, 6, 12, 24, 36, 48 and 72 h after surgery ('postoperative'). Data are reported as the mean \pm SE. $N = 10$ –16. Data were analyzed by analysis of variance with repeated measures followed by Tukey's procedures for pairwise comparisons. ND, no data.

^b $P < 0.001$, when compared with preoperative baseline value.

^c $P < 0.05$, when compared with preoperative baseline value.

^d $P < 0.01$, when compared with preoperative baseline value.

were inversely related to serum cortisol, serum prolactin, plasma ACTH and plasma β -endorphin (Fig. 1). Regression analysis revealed the following correlation coefficients: choline to cortisol, -0.642 ($P < 0.001$); choline to prolactin, -0.756 ($P < 0.001$); choline to ACTH, -0.458 ($P < 0.01$); and choline to β -endorphin, -0.726 ($P < 0.001$).

These results confirm and extend the previous finding that serum choline levels decrease during and after surgery [16].

The decline in serum choline concentrations could reflect a decreased entry of choline into the blood stream or its accelerated removal. The known sources of circulating choline are dietary phospholipids, hepatic synthesis of phosphatidylcholine, and presumably, the hydrolysis of acetylcholine and of membrane phospholipids. Our patients were in fasting during surgery and had no oral food intake for 12–24 h postoperatively. Also, most patients are fasted for at least 8 h before surgery. In humans, fasting serum choline concentrations are well maintained around 10 μ M without significant decrease unless dietary choline intake is restricted for 2–3 weeks [19]. Thus, it is unlikely that the 8–36 h absence of dietary choline in our patients affected the serum choline concentrations. Accelerated removal of choline from the circulation and/or decreased release of hepatic choline into the circulation may also explain the reduced serum choline levels.

Surgery is physiologically stressful and produces a variety of neuroendocrine and metabolic changes [4–7]. In fact, with surgery, stress hormones [5,6,9,10] such as cortisol, prolactin, ACTH and β -endorphin increase in the circulation (Table 1). Our finding that the decline in serum choline levels significantly correlates with an increase in serum cortisol, prolactin and plasma ACTH and β -endorphin is of interest. These observations raise the possibility that the decrease in serum choline levels results from the neuroendocrine–metabolic changes. This view is further supported by the preliminary data showing that cortisone (5 mg/kg) decreases the plasma choline levels in dogs by 40–60% within 60 min after the subcutaneous injection (Özarda İlçöl and Ulus, unpublished observation). Nevertheless, to determine a possible causal relation between the decline in serum choline and the observed increases in the stress hormones requires further investigation.

In summary, we have shown that the decrease in serum choline concentrations in patients undergoing surgery is associated with an increase in circulating cortisol, prolactin, ACTH and β -endorphin. These results will stimulate further research to elucidate the mechanisms involved in the alterations of serum choline concentrations during stress. The decline in serum choline could alter cholinergic neurotransmission and membrane phospholipid synthesis. Cholinergic neurons transmit signals for motor, preganglionic sympathetic fibers, and pre- and postganglionic parasympathetic fibers, as well as for neurons in the central nervous system involving memory, awareness, analgesia, temperature and blood pressure regulation. Restoring serum choline levels to normal by supplemental choline administration may

increase resistance to acute hemorrhage [1,15], enhance analgesia and relieve postoperative abdominal distension, gastric atony and retention and urinary retention. In addition, choline increases phospholipid synthesis [11] and has reticuloendothelial system stimulating activity [1], which might confer trauma protection, wound healing and tissue repair.

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- [1] Altura, B.M., Role of spleen in choline stimulation of reticuloendothelial system and resistance to acute hemorrhage, *Proc. Soc. Exp. Biol. Med.*, 158 (1978) 77–80.
- [2] Blusztajn, J.K., Choline, a vital amine, *Science*, 281 (1998) 794–795.
- [3] Buyukuysal, R.L., Ulus, I.H., Aydin, S. and Kiran, B.K., 3,4-Diaminopyridine and choline increase in vivo acetylcholine release in rat striatum, *Eur. J. Pharmacol.*, 281 (1995) 179–185.
- [4] Deuss, U., Dietrich, J., Kaulen, D., Frey, K., Spangenberg, W., Allolio, B., Matuszczak, M., Troidl, H. and Winkelmann, W., The stress response to laparoscopic cholecystectomy: investigation of endocrine parameters, *Endoscopy*, 26 (1994) 235–238.
- [5] Dubois, M., Pickar, D., Cohen, M.R., Roth, Y.F., Macnamara, T. and Bunney Jr., W.E., Surgical stress in humans is accompanied by an increase in plasma beta-endorphin immunoreactivity, *Life Sci.*, 29 (1981) 1249–1254.
- [6] Haxholdt, O.S.T., Kehlet, H. and Dyrberg, V., Effect of fentanyl on the cortisol and hyperglycemic response to abdominal surgery, *Acta Anaesthesiol. Scand.*, 25 (1981) 434–436.
- [7] Kehlet, H., Surgical stress response: does endoscopic surgery confer an advantage? *World J. Surg.*, 23 (1999) 801–807.
- [8] Köppen, A., Klein, J., Erb, C. and Löffelholz, K., Acetylcholine release and choline availability in rat hippocampus: effects of exogenous choline and nicotineamide, *J. Pharmacol. Exp. Ther.*, 282 (1997) 1139–1145.
- [9] Lacoumenta, S., Yeo, T.H., Burrin, J.M., Bloom, S.R., Paterson, J.L. and Hall, G.M., Fentanyl and the β -endorphin, ACTH and glucoregulatory hormonal response to surgery, *Br. J. Anaesth.*, 59 (1987) 713–720.
- [10] Noel, G.H., Sun, H.K., Stone, J.G. and Franz, A.G., Human prolactin and growth hormone release during surgery and other conditions of stress, *J. Clin. Endocrinol. Metab.*, 35 (1972) 840–846.
- [11] Savci, V. and Wurtman, R.J., Effect of cytidine on membrane phospholipid synthesis in rat striatal slices, *J. Neurochem.*, 64 (1995) 378–384.
- [12] Tucek, S., Short-term control of the synthesis of acetylcholine, *Prog. Biophys. Mol. Biol.*, 60 (1993) 59–69.
- [13] Ulus, I.H. and Wurtman, R.J., Choline administration: activation of tyrosine hydroxylase in dopaminergic neurons of rat brain, *Science*, 194 (1976) 1060–1061.
- [14] Ulus, I.H., Wurtman, R.J., Mauron, C. and Blusztajn, J.K., Choline increases acetylcholine release and protects against the stimulation-induced decrease in phosphatide levels within membranes of rat corpus striatum, *Brain Res.*, 484 (1989) 217–227.
- [15] Ulus, I.H., Arslan, B.Y., Savci, V. and Kiran, B.K., Restoration of blood pressure by choline treatment in rats made hypotensive by haemorrhage, *Br. J. Pharmacol.*, 116 (1995) 1911–1917.

- [16] Ulus, I.H., Ozyurt, G. and Korfali, E., Decreased serum choline concentrations in humans after surgery, childbirth and traumatic head injury, *Neurochem. Res.*, 23 (1998) 727–732.
- [17] Wang, F.L. and Haubrich, D.L., A simple, sensitive and specific assay for free choline in plasma, *Anal. Biochem.*, 63 (1975) 195–201.
- [18] Wurtman, R.J., Choline metabolism as a basis for the selective vulnerability of cholinergic neurons, *Trends Neurosci.*, 15 (1992) 117–122.
- [19] Zeisel, S.H., DaCosta, K.A., Franklin, P.D., Alexander, E.A., Lamont, J.T., Sheard, N.F. and Beiser, A., Choline, an essential nutrient for human, *FASEB J.*, 5 (1991) 2093–2098.