

Increased serum glial cell line-derived neurotrophic factor immunocontent during manic and depressive episodes in individuals with bipolar disorder

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Abstract

Glial cell line-derived neurotrophic factor (GDNF) is a neurotrophic factor from the transforming growth factor β family, which plays a role in the development and function of hippocampal cells. Preclinical studies suggest that changes in neurotrophic growth factor systems might be involved in the pathophysiology of mood disorders including bipolar disorder (BD) [E.J. Nestler, M. Barrot, R.J. DiLeone, A.J. Eisch, S.J. Gold, L.M. Monteggia, Neurobiology of depression, Neuron 34 (2002) 13–25]. This is the first study to analyze GDNF immunocontent in BD subjects across different mood states, including mania, depression, and remission (euthymia). Forty-four bipolar patients (14 depressed, 15 manic, and 15 euthymic) and 14 healthy controls, diagnosed according to the Structural Clinical Interview for DSM-IV were studied. Serum GDNF immunocontent was measured using Western blotting. Serum GDNF immunocontent was increased in manic ($F = 42.31$; $p = 0.001$; one-way ANOVA) and depressed ($F = 42.31$; $p = 0.004$; one-way ANOVA) bipolar patients, but not in euthymic patients as compared with controls. Our results indicate that changes in GDNF immunocontent occur during acute major affective episodes in bipolar subjects. These results further support the role of neurotrophins in the pathophysiology of bipolar disorder. Whether the observed increase in GDNF immunocontent correspond to a pathological or an adaptive response remains to be determined.

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Bipolar disorder (BD) is a prevalent, chronic, and life-threatening illness characterized by alternating episodes of mania and depression [41]. Postmortem studies showed abnormal density and size of neuronal and glial cells in several subregions of the prefrontal cortex, such as subgenual, orbitofrontal and dorsolateral prefrontal cortex in BD [10,26,33]. It has been

postulated that such morphological changes suggest impairment in cellular plasticity and resilience rather than a neurodegenerative pattern [32]. In addition, there is increasing evidence suggesting that neurotrophic signaling systems, which regulate cellular plasticity and survival, may be altered in bipolar disorder [16,25,36]. We have recently demonstrated that serum brain-derived neurotrophic factor (BDNF) is decreased in bipolar patients during acute manic and depressive episodes [11]. A recent study showed that glial cell line-derived neurotrophic factor (GDNF) is decreased in the whole blood of bipolar and

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unipolar subjects during “partial or full remission state” [38]. Further, preclinical studies demonstrated that the first-line mood stabilizers lithium and valproate increase BDNF and GDNF levels *in vivo* and *in vitro* [4,7,13].

GDNF is a neurotrophic factor belonging to the transforming growth factor β family, and was initially isolated based on its ability to induce dopamine uptake and cell survival in embryonic ventral midbrain cultures [22]. Although considered one of the most potent neurotrophic factors for dopaminergic neurons [6], GDNF is widely expressed throughout the brain [29], and exerts neuroprotective effects in several central and peripheral neuronal populations [1,6]. For instance, it has been demonstrated that GDNF regulates noradrenergic locus ceruleus-hippocampal pathway [30], and protects against kainate-induced oxidative stress in rat hippocampus [9]. Moreover, GDNF^{+/-} mutant mice present abnormal hippocampal synaptic transmission [24] and poorer spatial learning performance [14]. Considering recent reports that GDNF may play a role in the pathophysiology of BD [38] and in the effects of mood stabilizers, we investigated for the first time the serum GDNF immunocontent in BD patients across all mood states, including mania, depression, and remission (euthymia) as compared with matched healthy controls.

The sample consisted of 44 BD type I patients (14 depressed, 15 manic, and 15 euthymic) and 14 age- and gender-matched healthy volunteers. All patients were recruited from the Bipolar Disorders Program – Federal University of Rio Grande do Sul, Porto Alegre, Brazil, and the Inpatient Psychiatric Unit – Hospital Universitário de Santa Maria, Santa Maria, Brazil. Patients were 18 years or older (range 30–51 years), non-smokers, did not present active medical conditions and were not on any medication other from those prescribed for their psychiatric illness. BD diagnosis was carried out using the Structured Clinical Interview for DSM-IV—Axis I (SCID-I) [12], and manic and depressive symptoms were assessed using the Young Mania Rating Scale (YMRS) [42] and the 17-item Hamilton Depression Rating Scale (HDRS) [15], respectively. Bipolar subjects were considered manic, depressed or euthymic if they fulfilled criteria for current manic episode, current major depressive episode, or current in remission according to the SCID-I. The control group consisted of healthy individuals who manifested interest in participating in the study. Controls were 18 years or older (range 30–52 years), did not have history of major psychiatric disorders, as assessed by the SCID-I non-patient version, or history of dementia, mental retardation, cancer or tumor in their first-degree relatives. Control subjects were non-smokers and were not on medication. Five controls, six manic, and three depressed subjects overlap with the sample of our previous study [11]. This study was approved by the local ethics committee (Hospital de Clinicas de Porto Alegre, Porto Alegre, Brazil), and all subjects signed the informed consent before entering in the study. This study was conducted in accordance with the principles of Good Clinical Practice and the Declaration of Helsinki and its subsequent revisions.

Five milliliters of blood sample was collected by venipuncture without anticoagulants, and serum was obtained by centrifugation at 3000 g for 5 min and kept frozen at -80°C until the biochemical assay. Western blot analysis was per-

formed according to the following protocol: samples were diluted (1:4) with phosphate-buffered saline and then were dissolved (90 μg of protein/lane) in stopping solution (100 mM Tris-HCl, pH 8.8, 0.5 mM EDTA, 4% SDS, 4% glycerol, and 2% beta-mercaptoethanol). Protein samples were separated on 10% SDS-PAGE at 120 V and then electrotransferred at 15 V to PVDF membrane. Membranes were blocked with 5% albumin overnight at room temperature and then incubated for 1 h at room temperature with anti-human GDNF polyclonal (1:1000, Promega, Madison, USA). Blots were washed three times with tris-buffered saline (containing 0.5% Tween) and incubated with biotin-conjugated anti-chicken IgG (1:1000, Promega, Madison, USA) for 1 h at room temperature. Then blots were washed three times for 10 min each with 0.5% Tween-TBS and incubated with streptavidin (1:1000, Dako, Corporation A/S, Denmark) for 1 h at room temperature. The signal was detected by an enhanced chemiluminescence method (ECL kit, Amersham) and registered in a Kodak X-OMAT film. The intensity of the selected bands was analyzed using Optiquant program. Protein levels were quantified by Lowry's method, using bovine serum albumin as standard. Because peripheral GDNF levels are known to be altered in subjects with renal insufficiency [27,35] we assessed urea and creatinine levels in all subjects. Serum urea levels were assayed by urease-Berthelot reaction (Biodiagnostica, Paraná, Brazil) and measured at 600 nm. Serum creatinine levels were assayed by Jaffe reaction-picric acid alkaline (Biodiagnostica, Paraná, Brazil) and measured at 490 nm. Statistical analysis was performed using SPSS for Windows—version 12.0 (SPSS Inc., Chicago, USA). GDNF immunocontent of four groups (manic, depressed, euthymic, and controls) were compared using one-way ANOVA, and the individual differences were tested using Tukey HSD test if ANOVA was significant. Demographic characteristics between groups were compared using Chi-square and one-way ANOVA, and statistical significance was set at $p < 0.05$. Data are presented as percentage of control group.

Patients and controls did not differ in terms of age, gender, or education (all $p > 0.05$; see Table 1). Age of first episode and years of illness were not significantly different between depressed, manic, and euthymic bipolar patients. Serum GDNF immunocontent was higher in manic ($F = 42.31$; $p = 0.001$) and depressed ($F = 42.31$; $p = 0.004$) BD patients, as compared with euthymic patients and controls (see Fig. 1). All subjects had urea and creatinine levels within normal limits, and no differences in urea or creatinine levels were observed between groups (all $p > 0.05$), indicating that GDNF immunocontent was not altered by renal insufficiency.

Serum GDNF immunocontent was increased in bipolar patients during manic and depressive episodes and there was no difference between euthymic patients and healthy controls. This finding suggests that peripheral GDNF synthesis or release is increased during acute episodes in BD. Whether it represents a pathological or a compensatory mechanism remains to be determined. This result is in contrast to a recent report of lower whole blood GDNF levels assessed with ELISA in remitted bipolar and unipolar subjects [38]. Differences in sampling and immunoassay may account for this discrepancy. In addition, Takebayashi et al. [38] studied older subjects and did not control for possi-

Table 1
Demographics and clinical characteristics

	Control group (N = 14)	Bipolar patients			p-value
		Euthymic (N = 15)	Manic (N = 15)	Depressed (N = 14)	
Gender					
Male	31.2%	37.5%	56.3%	28.6%	0.123*
Female	68.8%	62.5%	43.8%	71.4%	
Age (years)	41.1 (11.1)	40.4 (10.3)	40.1 (9.3)	42.1 (8.2)	0.695**
Schooling (years)	7.9 (2.9)	9.6 (4.8)	8.3 (3.6)	8.5 (1.7)	0.101**
Age of first mood episode	–	21.3 (11.2)	27.1 (9.9)	22.0 (11.3)	0.410**
Years of illness	–	16.4 (12.0)	13.1 (8.3)	18.0 (14.1)	0.210**
HDRS	–	4.1 (2.1)	5.6 (2.9)	21.9 (2.36)	0.001**
YMRS	–	3.3 (3.4)	33.1 (4.1)	5.6 (3.1)	0.001**
Creatinine mg/dL ^a					
Male	0.90 (0.21)	1.10 (0.32)	1.13 (0.21)	1.24 (0.19)	0.233**
Female	0.78 (0.31)	0.81 (0.37)	1.19 (0.23)	0.99 (0.24)	0.455**
Urea mg/dL ^b	16.45 (1.24)	18.33 (2.55)	20.1 (1.44)	19.5 (1.44)	0.566**

(*) Chi-square test; (**) one-way ANOVA test; YMRS: Young Mania Rating Scale; HDRS: Hamilton Depression Rating Scale.

^a Serum reference levels → male: 0.6–1.5 mg/dL; female: 0.5–1.3 mg/dL.

^b Serum reference levels → 10–52 mg/dL.

ble effects of renal impairment, which is more common with increased age.

It has been demonstrated that bipolar patients have significant enlargement in the size of glial cells in specific cortical layers [33,40]. Moreover, it has been reported that manic patients have increased serum S100 β , an astrocytic neuroprotective protein [23]. It is known that neuronal injury may stimulate glial activity, and separation of astroglial cells from cortical neurons was shown to promote neuronal death [31]. GDNF supports the survival of dopaminergic [21] and noradrenergic [5] brain cells *in vivo*, as well as peripheral motor [17] and sensory neurons [39]. In peripheral blood, studies showed that bipolar patients have increased malondialdehyde (a marker of lipid peroxidation) [20,28], and decreased catalase [28,34], indicating an increased oxidative stress status. Using single cell gel electrophoresis assay, we recently found increased DNA fragmentation in peripheral blood in BD, possible due to increased oxidative stress [3]. Chao and Lee [8] demonstrated that sub-

chronic infusion of recombinant human GDNF increased superoxide dismutase, catalase, and glutathione peroxidase activity in rat striatum, suggesting that GDNF may exert antioxidant properties. Interestingly, Sawada et al. [36] showed that preincubation with GDNF blocked the DNA cleavage induced by bleomycin sulphate and L-buthionine-[S,R]-sulfoximine exposure in cultured mesencephalic neurons. Although speculative, it is possible that the increment in GDNF immunocontent during acute manic and depressive episodes might be an adaptative response against oxidative stress.

Some limitations must be taken in consideration. In the present study, all bipolar patients were on medication, thus we cannot exclude that the GDNF immunocontent may be influenced by the treatment. Although a previous study found no differences in GDNF levels between bipolar patients with or without lithium/antidepressant therapy [38], preclinical studies demonstrated that GDNF levels may be altered by antidepressants [18], antipsychotics [37], and mood stabilizers [4,7]. Studies conducted in medication-free subjects are warranted to further clarify this issue. Second, because we measured GDNF immunocontent in the serum, we cannot determine whether these peripheral changes reflect actual changes in the central nervous system. Previous data indicate that GDNF penetrates very poorly across the brain blood barrier [2,19].

In conclusion, serum GDNF immunocontent is increased in bipolar patients during acute manic and depressive episodes. Whether it represents a pathological or a compensatory response remains to be determined. This finding further supports that bipolar disorder is associated with multiple changes in neurotrophic signaling systems.

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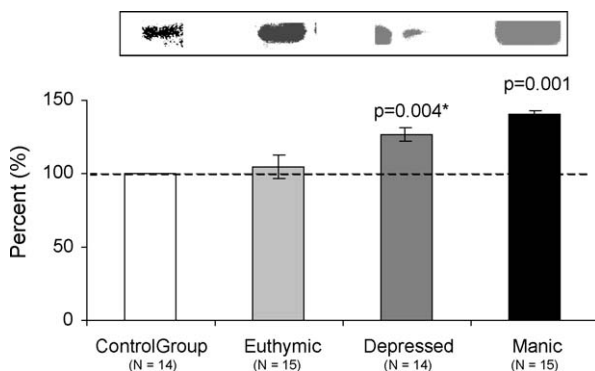


Fig. 1. Serum GDNF content in BD patients and controls. Serum content of GDNF was measured by immunoblotting applying 90 μ g of protein per lane. Values of the GDNF were expressed in percentages assuming control sample as being 100% (mean \pm S.E.M.). Inset is a representative immunoblot from one of the subjects.

References

- [1] M. Airaksinen, M. Saarma, The GDNF family: signaling, biological functions and therapeutic value, *Nat. Rev. Neurosci.* 3 (2002) 383–394.
- [2] D.S. Albeck, B.J. Hoffer, D. Quissell, L.A. Sanders, G. Zerbe, A.C. Granholm, A non-invasive transport system for GDNF across the blood-brain barrier, *Neuroreport* 7 (8) (1997) 2293–2298.
- [3] A.C. Andrezza, B.N. Frey, B. Erdtmann, M. Salvador, F. Rombaldi, A. Santin, C.A. Gonçalves, F. Kapczinski, DNA damage in bipolar disorder, *Psychiatry Res.*, in press.
- [4] F. Angelucci, L. Aloe, P. Jimenez-Vasquez, A.A. Mathe, Lithium treatment alters brain concentrations of nerve growth factor, brain-derived neurotrophic factor and glial cell line-derived neurotrophic factor in a rat model of depression, *Int. J. Neuropsychopharmacol.* 6 (2003) 225–231.
- [5] E. Arenas, M. Trupp, P. Akerud, C.F. Ibanez, GDNF prevents degeneration and promotes the phenotype of brain noradrenergic neurons in vivo, *Neuron* 15 (1995) 1465–1473.
- [6] M.C. Bohn, B. Connor, D.A. Kozlowski, M.H. Mohajeri, Gene transfer for neuroprotection in animal models of Parkinson's disease and amyotrophic lateral sclerosis, *Novartis Found. Symp.* 231 (2000) 70–89, discussion 89–93.
- [7] L.M. Castro, M. Gallant, L.P. Niles, Novel targets for valproic acid: up-regulation of melatonin receptors and neurotrophic factors in C6 glioma cells, *J. Neurochem.* 95 (2005) 1227–1236.
- [8] C.C. Chao, E.H. Lee, Neuroprotective mechanism of glial cell line-derived neurotrophic factor on dopamine neurons: role of antioxidation, *Neuropharmacology* 38 (1999) 913–916.
- [9] H. Cheng, Y.S. Fu, J.W. Guo, Ability of GDNF to diminish free radical production leads to protection against kainite-induced excitotoxicity in hippocampus, *Hippocampus* 14 (2004) 77–86.
- [10] D. Cotter, L. Hudson, S. Landau, Evidence for orbitofrontal pathology in bipolar disorder and major depression, but not in schizophrenia, *Bipolar Disord.* 7 (2005) 358–369.
- [11] A.B. Cunha, B.N. Frey, A.C. Andrezza, J.D. Goi, A.R. Rosa, C.A. Gonçalves, A. Santin, F. Kapczinski, Serum brain-derived neurotrophic factor is decreased in bipolar disorder during depressive and manic episodes, *Neurosci. Lett.* 8 (2006) 215–219.
- [12] M.B. First, R.L. Spitzer, M. Gibbon, J.B. Williams, Structured Clinical Interview for DSM-IV (SCID-I), Biomedics Research Department, New York, 1998.
- [13] B.N. Frey, A.C. Andrezza, K.M. Ceresér, M.R. Martins, S.S. Valvassori, G.Z. Réus, J. Quevedo, F. Kapczinski, Effects of mood stabilizers on hippocampus BDNF levels in an animal model of mania, *Life Sci.* 13 (2006) 281–286.
- [14] R. Gerlai, A. McNamara, D.L. Choi-Lundberg, M. Armanini, J. Ross, L. Powell-Braxton, H.S. Phillips, Impaired water maze learning performance without altered dopaminergic function in mice heterozygous for the GDNF mutation, *Eur. J. Neurosci.* 14 (2001) 1153–1163.
- [15] M. Hamilton, A rating scale for depression, *J. Neurol. Neurosurg. Psychiatry* 23 (1960) 56–62.
- [16] K. Hashimoto, E. Shimizu, M. Iyo, Critical role of brain-derived neurotrophic factor in mood disorders, *Brain Res. Brain Res. Rev.* 45 (2004) 104–114.
- [17] C.E. Henderson, H.S. Phillips, R.A. Pollock, A.M. Davies, C. Lemeulle, M. Armanini, L. Simmons, B. Moffet, R.A. Vandlen, L.C. Simpson, GDNF: a potent survival factor for motoneurons present in peripheral nerve and muscle, *Science* 266 (1994) 1062–1064.
- [18] K. Hisaoka, A. Nishida, T. Koda, M. Miyata, H. Zensho, S. Morinobu, M. Ohta, W. Yamawaki, Antidepressant drug treatments induce glial cell line-derived neurotrophic factor (GDNF) synthesis and release in rat C6 glioblastoma cells, *J. Neurochem.* 79 (2001) 25–34.
- [19] A.J. Kastin, V. Akerstrom, W. Pan, Glial cell line-derived neurotrophic factor does not enter normal mouse brain, *Neurosci. Lett.* 17 (2003) 239–241.
- [20] M. Kuloglu, B. Ustundag, M. Atmaca, H. Canatan, A.E. Tezcan, N. Cinkilinc, Lipid peroxidation and antioxidant enzyme levels in patients with schizophrenia and bipolar disorder, *Cell Biochem. Funct.* 20 (2002) 171–175.
- [21] P.A. Lapchak, P.J. Miller, F. Collins, S. Jiao, Glial cell line-derived neurotrophic factor attenuates behavioural deficits and regulates nigrostriatal dopaminergic and peptidergic markers in 6-hydroxydopamine-lesioned adult rats: comparison of intraventricular and intranigral delivery, *Neuroscience* 78 (1997) 61–72.
- [22] L.F. Lin, D.H. Doherty, J.D. Lile, S. Bektesh, F. Collins, GDNF: a glial cell line-derived neurotrophic factor for midbrain dopaminergic neurons, *Science* 21 (1993) 1130–1132.
- [23] R. Machado-Vieira, D.R. Lara, L.V. Portela, C.A. Gonçalves, J.C. Soares, F. Kapczinski, D.O. Souza, Elevated serum S100B protein in drug-free bipolar patients during first manic episode: a pilot study, *Eur. Neuropsychopharmacol.* 12 (2002) 269–272.
- [24] A. Nanobashvili, M.S. Airaksinen, M. Kokaia, J. Rossi, F. Asztely, K. Olofsson, P. Mohapel, M. Saarma, O. Lindvall, Z. Kokaia, Development and persistence of kindling epilepsy are impaired in mice lacking glial cell line-derived neurotrophic factor family receptor alpha 2, *Proc. Natl. Acad. Sci. U.S.A.* 24 (2000) 12312–12317.
- [25] E.J. Nestler, M. Barrot, R.J. DiLeone, A.J. Eisch, S.J. Gold, L.M. Monteggia, Neurobiology of depression, *Neuron* 34 (2002) 13–25.
- [26] D. Ongur, W.C. Drevets, J.L. Price, Glial reduction in the subgenual prefrontal cortex in mood disorders, *Proc. Natl. Acad. Sci. U.S.A.* 27 (1998) 13290–13295.
- [27] H. Onodera, T. Nagata, M. Kanazawa, Y. Taguma, Y. Itoyama, Increased plasma GDNF levels in patients with chronic renal diseases, *Nephrol. Dial. Transplant.* 14 (1999) 1604–1605.
- [28] M.E. Ozcan, M. Gulec, E. Ozerol, R. Polat, O. Akyol, Antioxidant enzyme activities and oxidative stress in affective disorders, *Int. Clin. Psychopharmacol.* 19 (2004) 89–95.
- [29] N.A. Pochon, A. Menoud, J.L. Tseng, A.D. Zurn, P. Aebischer, Neuronal GDNF expression in the adult rat nervous system identified by in situ hybridization, *Eur. J. Neurosci.* 9 (1997) 463–471.
- [30] E.M. Quintero, L.M. Willis, V. Zaman, J. Lee, H.A. Boger, A. Tomac, B.J. Hoffer, I. Stromberg, A.C. Granholm, Glial cell line-derived neurotrophic factor is essential for neuronal survival in the locus ceruleus-hippocampal noradrenergic pathway, *Neuroscience* 124 (2004) 137–146.
- [31] G. Rajkowska, Postmortem studies in mood disorders indicate altered numbers of neurons and glial cells, *Biol. Psychiatry* 48 (2000) 766–777.
- [32] G. Rajkowska, Depression: what we can learn from postmortem studies, *Neuroscientist* 9 (2003) 273–284.
- [33] G. Rajkowska, A. Halaris, L.D. Selemon, Reductions in neuronal and glial density characterize the dorsolateral prefrontal cortex in bipolar disorder, *Biol. Psychiatry* 49 (741) (2001) 752.
- [34] P.K. Ranjekar, A. Hinge, M.V. Hegde, M. Ghate, A. Kale, S. Sitasawad, U.V. Wagh, V.B. Debsikdar, S.P. Mahadik, Decreased antioxidant enzymes and membrane essential polyunsaturated fatty acids in schizophrenic and bipolar mood disorder patients, *Psychiatry Res.* 1 (2003) 109–122.
- [35] M. Saarma, GDNF—a stranger in the TGF-beta superfamily? *Eur. J. Biochem.* 267 (2000) 6968–6971.
- [36] H. Sawada, M. Ibi, T. Kihara, M. Urushitani, M. Nakanishi, A. Akaike, S. Shimohama, Neuroprotective mechanism of glial cell line-derived neurotrophic factor in mesencephalic neurons, *J. Neurochem.* 74 (2000) 1175–1184.
- [37] Z. Shao, L.E. Dyck, H. Wang, X.M. Li, Antipsychotic drugs cause glial cell line-derived neurotrophic factor secretion from C6 glioma cells, *J. Psychiatry Neurosci.* 31 (2006) 32–37.
- [38] M. Takebayashi, K. Hisaoka, A. Nishida, M. Tsuchioka, I. Miyoshi, T. Kozuru, S. Hikasa, Y. Okamoto, H. Shinno, S. Morinobu, S. Yamawaki, Decreased levels of whole blood glial cell line-derived neurotrophic factor (GDNF) in remitted patients with mood disorders, *Int. J. Neuropsychopharmacol.* 28 (2005) 1–6.
- [39] M. Trupp, M. Ryden, H. Jornvall, H. Funakoshi, T. Timmusk, E. Arenas, C.F. Ibanez, Peripheral expression and biological activities of GDNF, a new

- neurotrophic factor for avian and mammalian peripheral neurons, *J. Cell Biol.* 130 (1995) 137–148.
- [40] N. Uranova, D. Orlovskaya, O. Vikhreva, I. Zimina, N. Kolomeets, V. Vostrikov, V. Rachmanova, Electron microscopy of oligodendroglia in severe mental illness, *Brain Res. Bull.* 55 (2001) 597–610.
- [41] E. Vieta, Mood stabilization in the treatment of bipolar disorder: focus on quetiapine, *Hum. Psychopharmacol.* 20 (2005) 225–236.
- [42] R.C. Young, J.T. Biggs, V.E. Ziegler, D.A. Meyer, A rating scale for mania: reliability, validity, and sensitivity, *Br. J. Psychiatry* 133 (1978) 429–435.