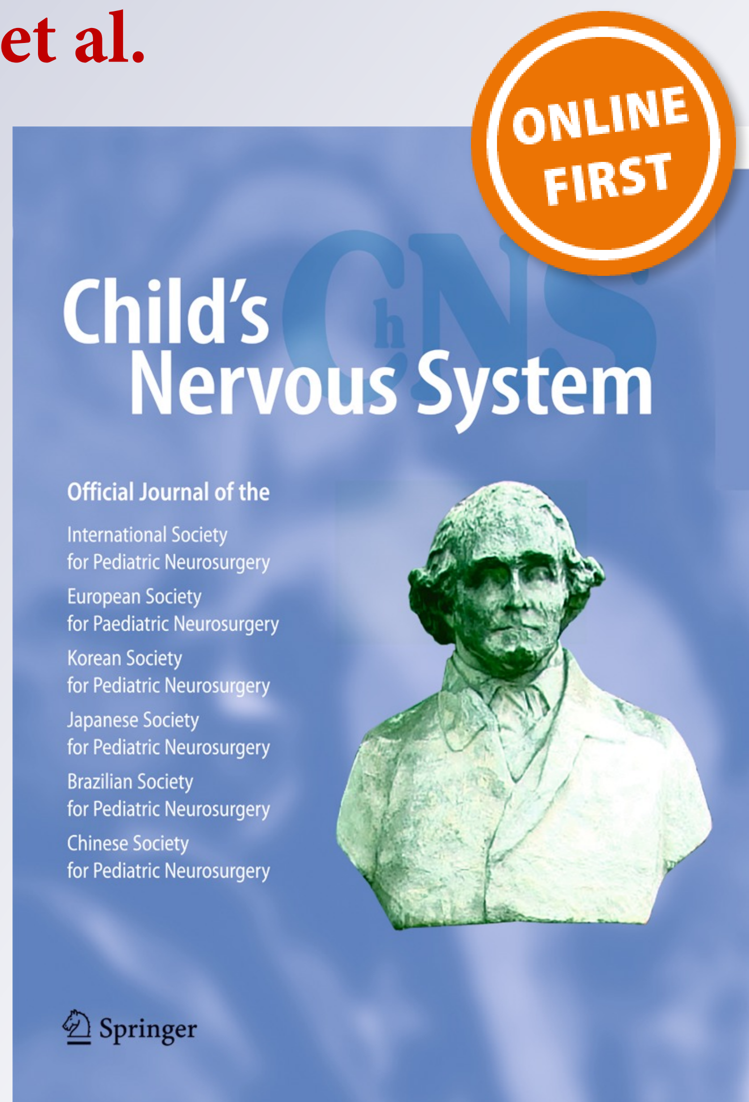
Extensive miRNA expression analysis in craniopharyngiomas

Jill Samis, Elio F. Vanin, Simone Treiger Sredni, Maria de Fátima de Bonaldo, Fabricio F. Costa, Tadanori Tomita, Reema Habiby, et al.

Child's Nervous System

ISSN 0256-7040

Childs Nerv Syst
DOI 10.1007/s00381-016-3131-1



Springer

Your article is protected by copyright and all rights are held exclusively by Springer-Verlag Berlin Heidelberg. This e-offprint is for personal use only and shall not be self-archived in electronic repositories. If you wish to self-archive your article, please use the accepted manuscript version for posting on your own website. You may further deposit the accepted manuscript version in any repository, provided it is only made publicly available 12 months after official publication or later and provided acknowledgement is given to the original source of publication and a link is inserted to the published article on Springer's website. The link must be accompanied by the following text: "The final publication is available at link.springer.com".

Springer

Author's personal copy

Childs Nerv Syst
DOI 10.1007/s00381-016-3131-1

ORIGINAL PAPER

Extensive miRNA expression analysis in craniopharyngiomas

Jill Samis[1,2] · **Elio F. Vanin**[3] · **Simone Treiger Sredni**[2,3,4] · **Maria de Fátima de Bonaldo**[3] · **Fabricio F. Costa**[3] · **Tadanori Tomita**[2,4] · **Reema Habiby**[1,2] · **Donald Zimmerman**[1,2] · **Marcelo B. Soares**[3,4,5]

Received: 20 May 2016 / Accepted: 26 May 2016
© Springer-Verlag Berlin Heidelberg 2016

Abstract
Purpose Craniopharyngiomas are benign tumors of the sellar or parasellar regions. They arise from the remnants of Rathke's pouch and are considered a "developmental disease." microRNAs are short non-coding RNAs that play a key regulatory role in the control of expression of entire gene networks. We performed an extensive analysis of miRNAs in craniopharyngiomas aiming to identify a miRNA expression signature that might aid in the prognosis of disease progression and outcome.
Methods Thirty-seven craniopharyngioma samples from twenty-three patients, ten age-matched controls from autopsy, and ten infant controls from the developing pituitary from autopsy were evaluated for the expression of 754 miRNAs using TaqMan® Low Density Arrays (TLDAs) v2.0 (Applied Biosystems, Foster City, CA).
Results Among the most differentially expressed miRNAs, downregulation of miR-132 appears to be a marker of aggressiveness and also plays a role in epithelial–mesenchymal transition.
Conclusions This is the first time that an extensive study of miRNA expression has been performed in craniopharyngiomas. Further research needs to be performed to investigate the potential role of miR-132 in the development and progression of craniopharyngiomas, and its value as a prognostic marker of aggressiveness.

Keywords Craniopharyngiomas · microRNA · miRNA · miR-132 · miR-596

In Memoriam of Elio F. Vanin

Jill Samis and Elio F. Vanin contributed equally to this work.

✉ Simone Treiger Sredni
ssredni@luriechildrens.org

[1] Division of Endocrinology, Ann and Robert H. Lurie Children's Hospital of Chicago, 225 E Chicago Ave., Chicago, IL 60611, USA

[2] Feinberg School of Medicine, Northwestern University, 303 E Chicago Ave, Chicago, IL 60611, USA

[3] Cancer Biology and Epigenomics, Stanley Manne Children's Research Institute, 2430 North Halsted St., Chicago, IL 60614, USA

[4] Division of Pediatric Neurosurgery, Ann and Robert H. Lurie Children's Hospital of Chicago, 225 E Chicago Ave., Chicago, IL 60611, USA

[5] Present address: University of Illinois College of Medicine at Peoria, One Illini Drive, Peoria, IL 61605, USA

Introduction

Craniopharyngiomas are benign tumors of the sellar or parasellar regions that, although histologically classified as benign, frequently invade the surrounding vital brain structures leading to a multitude of consequences, such as pituitary hormone deficits, hydrocephalus, and vision impairments [1]. Although relatively rare, with an estimated 350 diagnoses in the USA per year, craniopharyngiomas make up 5.6–15 % of all pediatric brain tumors, making them the third most common childhood intracranial tumor [1]. The current therapy for craniopharyngiomas is surgical excision, either gross total resection or sub-total resection, with or without cranial irradiation. Although surgery can sometimes result in cure, it can lead to permanent damage due to disruption of the vital surrounding structures, even in the hands of the most experienced surgeons. Variable recurrence rates are reported following surgery, both with gross total resection and sub-total resection. If the biological basis and clinical behavior of the tumor were known preoperatively, surgical planning for gross total

Published online: 07 June 2016

Author's personal copy

resection versus a sub-total resection and informed treatment decision for cranial irradiation could be made, thereby improving the outcomes and quality of life for patients.

Craniopharyngiomas are considered a "developmental disease" as they arise from the remnants of Rathke's pouch, which is a structure in the developing embryo arising from the roof of the mouth that becomes part of the anterior pituitary gland prior to birth [1]. The development of the pituitary gland has been extensively studied in mouse models. Multiple factors and signaling pathways have been shown to play an important role, both in the formation and maintenance of the gland. Changes in these factors have led to many defects, including improper formation of the pituitary or hypothalamus, isolated hormone deficiencies, and malformation of the structures of the eye and optic nerve [2, 3]. While many studies have been performed investigating expression levels of genes involved in these processes, a cause for the development of craniopharyngiomas remains unknown.

Epigenetics refers to the study of chemical modifications that occur in the DNA and histone tails and/or alterations in non-coding RNAs that may affect gene expression. Albeit not encoded in the DNA sequence, epigenetic alterations are heritable [4]. These changes have been shown to be responsible for many types of cancer. Among the various epigenetic changes that have been observed, changes in non-coding RNAs, especially small, non-coding RNAs such as small interfering RNAs (siRNAs) and microRNAs (miRNAs) have been reported. miRNAs are short non-coding RNAs of approximately 20 nucleotides in length, which negatively regulate gene expression by causing degradation of complementary RNAs or by blocking messenger RNA translation, ultimately having a major impact in the regulation of protein-coding gene networks and pathways [3]. They are known as master regulators of gene expression, and they have been associated with a wide range of biological processes, including differentiation, proliferation, and apoptosis [5, 6]. Alterations in the expression of miRNAs and their subsequent effect on protein coding regions have implicated miRNAs in multiple human diseases, such as brain tumors, heart diseases, autoimmune diseases, and cancer [6, 7]. miRNA expression profiles have been studied in many cancers, such as colon, kidney, prostate, urinary bladder, lung, and breast cancers and pediatric brain tumors [8–10] among others. The oncogenic and tumor-suppressive functions of several miRNAs have been characterized, and many have been described as potential therapeutic targets [6].

To date, there has been only one study that conducted extensive analysis of the miRNA expression profile in craniopharyngiomas, which concentrated on the miRNAs that targeted the Wnt pathway [11]. In this study, we investigated the expression of 754 miRNAs in craniopharyngiomas aiming to identify a miRNA expression signature that might aid in the prognosis of disease progression and outcome. To the best of our knowledge, this is the first comprehensive analysis of miRNA expression profiles in craniopharyngiomas.

Materials and methods

Patients and samples

Thirty-four craniopharyngioma samples from twenty-three patients, ten age-matched controls from autopsy, and ten infant controls from the developing pituitary from autopsy were collected retrospectively from the Pathology Department at the Lurie Children's Hospital at Chicago, IL (formerly Children's Memorial Hospital), under an IRB-approved protocol. Formalin-fixed, paraffin-embedded (FFPE) samples of craniopharyngiomas were obtained from pediatric patients treated between 1 January 1980 and 22 September 2010. The tumor information, as well as the clinicopathologic information such as age of the patient at the time the samples were obtained, sex, initial treatment modality, whether the tumors recurred, and treatment of the time of the recurrence were collected by a retrospective review of the medical records and are shown in Table 1.

RNA extraction and quantification of miRNA expression

Total RNA from thick cores of the FFPE samples was isolated using the Ambion RecoverAll kit (Ambion/Applied Biosystems, Foster City, CA) according to the manufacturer's instructions. The amount and quality of the RNA was determined by the absorbance of the RNA at 260 nm. Reverse transcriptions were performed using TaqMan® microRNA Reverse Transcription Kit (Life Technologies), according to the manufacturer's instructions. The cDNA was amplified before running the TaqMan® Low Density Arrays (TLDAs) v2.0 plates. This was done using the TaqMan® PreAmp Master Mix (Life Technologies, cat. no. 4391128) and the Megaplex™ PreAmp Primers, Human Pool Set v3.0 (cat. no. 4444748, Life Technologies). TaqMan® Low Density Arrays (TLDAs) v2.0 (Applied Biosystems, Foster City, CA) were then used to quantify the expression levels of 754 mature miRNAs and 3 controls (1 RNU44, 1 RNU48, and 4 MamU6 per plate) distributed in two separate plates.

Hierarchical clustering, statistical analysis, and chromosomal localization

Hierarchical clustering was performed using MeV [12], a desktop application for the analysis, visualization, and data mining of large-scale genomic data. Pearson correlation coefficient method with average linkage was applied. Comparisons to determine the significant differentially expressed miRNAs among the various clustering groups were

Author's personal copy

Table 1 Craniopharyngioma cohort description

Patient	Event	Age	Gender	Initial treatment modality	Recurrence	Clustering group	Classification
1	A	3 years 5 months	M	Surgery (GTR)	Yes	2	PR
2	A	6 years 6 months	F	Surgery (GTR)	Yes	2	PR
	B	8 years 4 months		Surgery	Yes	2	R
4	B	5 years 6 months	N.A.	N.A.	Yes	2	R
	C	10 years 1 month		N.A.	Yes	2	R
	D	12 years 4 months		N.A.	Yes	2	R
	E	13 years 8 months		N.A.	Yes	None	R
5	A	5 years 8 months	F	Surgery (GTR, Ommaya)	Yes	3	PR
6	A	6 years 5 months	M	Surgery	Yes	None	PR
	B	15 years 0 month		N.A.	No	3	R
7	A	15 months	F	Surgery (GTR)	No	3	PNR
8	A	7 years 4 months	M	Surgery (STR)	Yes	3	PR
	B	10 years 2 months		N.A.	Yes	3	R
9	B	5 years 8 months	N.A.	N.A.	Yes	3	R
10	B	8 years 9 months	N.A.	N.A.	Yes	3	R
11	A	10 years 8 months	M	Surgery, XRT	Yes	3	PR
12	A	12 years 1 month	M	Surgery (STR), XRT	Yes	3	PR
	B	13 years 2 months		N.A.	No	3	R
13	A	10 years 1 month	M	Surgery (GTR)	Yes	3	PR
14	A	6 years 11 month	M	Surgery (GTR)	Yes	3	PR
15	A	6 years 0 month	F	Surgery (GTR)	No	3	PNR
16	A	6 years 5 months	F	Surgery	No	3	PNR
17	A	16 years 4 months	F	Surgery (GTR)	No	3	PNR
18	A	16 years 1 month	F	Surgery (STR)	Yes	3	PR
19	A	4 years 4 months	M	Surgery (STR)	Yes	3	PR
20	A	2 years 6 months	M	Surgery (GTR)	Yes	1	PR
	B	2 years 11 months		N.A.	Yes	1	R
	D	9 years 9 months		N.A.	No	1	R
21	A	16 years 2 months	M	Surgery	Yes	1	PR
	B			N.A.	Yes	1	R
22	A	5 years 9 months	M	Surgery (GTR)	Yes	1	PR
	B	10 years 8 months		N.A.	No	1	R
23	A	11 years 7 months	M	Surgery (STR)	Yes	1	PR
	B			N.A.	Yes	1	R

Abbreviations: *A* primary tumor, *B* first relapse, *C* second relapse, *D* third relapse, *XRT* radiation therapy, *GTR* gross total resection, *STR* subtotal resection, *R* relapse, *PR* primary tumor that relapsed, *PNR* primary tumor that did not relapse, *N.A.* information not available

performed using RealTime StatMiner® (Integromics®). For all the comparisons, a cycle threshold (Ct) value greater than 35 was considered to be the absence of expression; MammU6 was used as the endogenous control. The StaMiner program used the empirical Bayes moderated *t* test to generate the ΔΔCt values for each comparison, and the Benjamini-Hochberg method was used to calculate the false discovery rate (FDR). StatMiner® was used to generate an excel table with *p* values and fold changes. Then, using this table, all the miRNAs with a *p* value of 0.05 or greater were excluded and the most differentially expressed miRNAs were determined. Chromosomal localization was determined by searching Human Feb. 2009 (GRCh37/hg19) assembly version of the human genome in the UCSC Genome browser.

Results and discussion

Characteristics of the patients and samples

A total of 37 FFPE craniopharyngioma samples from 23 patients and 20 control pituitary gland tissue (ten from infants

Author's personal copy

and ten age-matched controls) were used to evaluate miRNA expression. Of the 37 craniopharyngioma samples, 20 were primary tumors, 4 of these did not relapse (primary non-relapse (PNRs)), and 16 relapsed (primary relapse (PRs)). The remaining 17 tumor samples were relapses. Three craniopharyngioma samples (4A, 8C, and 20C) did not pass the quality control criteria and were excluded from the study (Table 1).

Hierarchical clustering of tumor and control samples

Primary tumors (PNRs and PRs), controls (infant and age-matched), and relapses were submitted to unsupervised hierarchical clustering using Pearson correlation with average linkage (Fig. 1). The clustering result showed that most of the tumors cluster into three clustering groups (groups 1, 2, and 3). Samples from patients 20, 21, 22, and 23 (primary and relapses) cluster in group 1, samples from patient 4 (primary and relapses) cluster in group 2, and samples from patients 8 and 12 (primary and relapse) cluster in group 3, together with all four primary tumors that did not relapse (PNRs—7, 15, 16, and 17). For this reason, group 3 was interpreted as a "lower aggressiveness group." All of the controls clustered together.

Two craniopharyngiomas, 4E and 6A, did not cluster with any group (indicated by the blue line). We hypothesize that this is due to previous treatments including four surgical excisions and radiation therapy, which may have been altered its expression profile. Two cases have been reported in which a craniopharyngioma have become a different tumor after radiation therapy, one was converted to an astrocytoma [13] and the other into a meningioma [14]. Sample 6A did not cluster with the other craniopharyngioma samples but clustered with the controls, which is most probably due to contamination by normal pituitary tissue. Both samples (4E and 6A) were excluded from further the analyses.

Comparison of microRNA expression profiles of craniopharyngioma samples and controls

Craniopharyngioma samples were compared with the controls. Based on fold changes and p values, a list of the most differentially expressed miRNAs was developed (Table 2). High fold changes are observed due to cDNA pre-amplification as described in the "Material and methods" section.

From the miRNAs presented in Table 2, only 6 of the 30 miRNAs were upregulated in craniopharyngiomas. The remaining 24 miRNAs were downregulated. Upon investigating the chromosomal location of the most significantly differentially expressed miRNAs, we observed that 9 of the 24 most downregulated miRNAs are located on chromosome X (miR-503**, miR-509-5p, miR-424**, miR-450b-5p**, miR-508-3p, miR-424#**, miR-542-3p**, miR-542-5p**, and miR-767-3p); 6 of them (miR-503**, miR-424**, 542-3p, 542-5p, miR-424#**, and miR- 450b-5p) were found to be in close proximity to chromosome X (q26.3). Interestingly, micro duplication of this locus has been reported to lead to a growth hormone/prolactin-secreting pituitary adenoma [15, 16]. The upregulated miRNAs are randomly distributed in the genome.

All the miRNAs in Table 2, except miR-450b-5p**, have been associated with cancer [17–39]. The miR-1274b has been shown to be completely contained within $tRNA^{Lys5}$ molecule [40]. miRNA-1260, which was upregulated in tumors, has been found to be upregulated when primary cutaneous malignant melanomas and cutaneous malignant melanoma metastases were compared to benign melanocytic nevi [41]. Interestingly, miR-1260 is located within the 3′ untranslated region (opposite orientation) of the neuroglobin gene, which is involved in increasing oxygen availability and providing protection under hypoxic/ischemic conditions [42].

We found miR-767-3p to be the most downregulated miRNA. miR-767-3p has been found to downregulate MGMT (O^6-methylguanine-DNA methyltransferase) in human glioblastomas [39]. Altered expression of miR-200b* (200b-5p) has been reported to occur in patients affected by lung adenocarcinomas, lung granulomas, and in healthy smokers [43]. On the other hand, miR-542-3p and the closely linked miR-542-5p both have been demonstrated to have tumor suppressor function in neuroblastoma [44, 45]. For miR-375, there have been many reports showing that it is downregulated in tumors, including in colorectal cancer [46], laryngeal squamous cell carcinoma [47], and esophageal squamous cell carcinoma [48, 49]. On the other hand, miR-1225-3p has been reported to be overexpressed on sporadic recurring pheochromocytomas and multiple endocrine neoplasia type 2 [36].

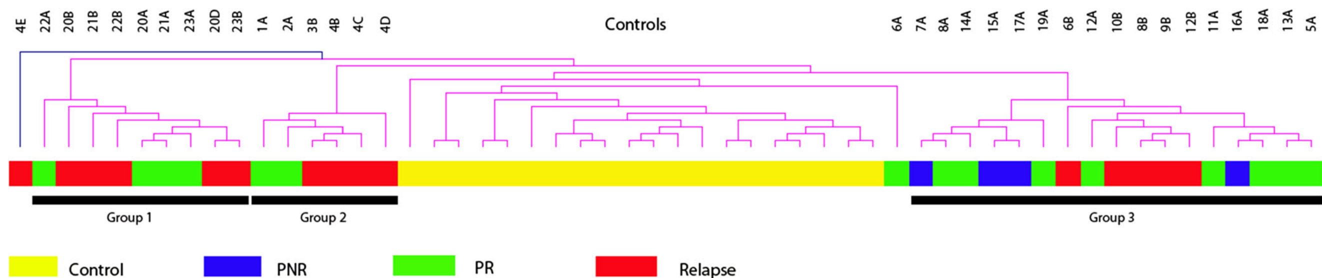


Fig. 1 Unsupervised hierarchical clustering showing the three clustering groups and controls

Author's personal copy

Table 2 List of the most differentially expressed microRNAs when comparing craniopharyngiomas and controls

miRNA	Fold change	p value	Chromosomal site	References
miR-1274b	435,680.70	1.31×10^{-21}	Chr.19(q13,41)	40
miR-1260	64,249.30	1.5×10^{-19}	Chr. 14(q24.3)	41, 42
miR-549	2514.10	2.4×10^{-27}	Chr. 15(q25.1)	17
miR-1183	2482.00	1.7×10^{-15}	Chr. 7(p15.3)	18
miR-10b#	2187.00	8.0×10^{-21}	Chr. 2(q31.1)	19
miR-205	185.9	2.2×10^{-13}	Chr. 1(q32.2)	20
miR-135b	−16.6	9.1×10^{-13}	Chr. 1(q32.1)	21
miR-539*	−86.8	3.5×10^{-13}	Chr. 14(q32.31)	22
miR-544*	−105.8	4.0×10^{-13}	Chr. 14(q32.31)	23
miR-410*	−156.4	3.2×10^{-14}	Chr. 14(q32.31)	24
miR-323-3p*	−250.5	3.5×10^{-13}	Chr. 14(q32.31)	25
miR-132#	−525.8	1.9×10^{-16}	Chr. 17(p13.3)	52, 53, 54, 55, 56
miR-411#*	−554.3	3.7×10^{-16}	Chr. 14(q32.31)	26
miR-503**	−591.2	2.3×10^{-21}	Chr. X(q26.3)	27
miR-509-5p	−872.1	3.9×10^{-15}	Chr. X(q27.3)	28
miR-770-5p	−948.2	2.5×10^{-13}	Chr. 14(q32.2)	29
miR-424**	−1078.90	1.2×10^{-15}	Chr. X(q26.3)	30
miR-450b-5p**	−1145.50	1.2×10^{-15}	Chr. X(q26.3)	None
miR-508-3p	−1218.50	5.3×10^{-14}	Chr. X(q27.3)	31
miR-29b-2#	−1830.50	1.4×10^{-21}	Chr. 1(q32.2)	32
miR-338-3p	−1944.50	1.2×10^{-15}	Chr. 17(q25.3)	33
miR-889*	−2678.70	7.4×10^{-14}	Chr. 14(q32.31)	34
miR-424#**	−3212.70	4.3×10^{-28}	Chr. X(q26.3)	30
miR-542-3p**	−5143.10	8.1×10^{-22}	Chr. Xq(26.3)	35
miR-375	−6312.80	3.4×10^{-15}	Chr. 2(q35)	46, 47, 48, 49
miR-1225-3p	−7103.70	1.3×10^{-21}	Chr. 16(p13.3)	36
miR-542-5p**	−9837.20	1.2×10^{-20}	Chr. X(q26.3)	45
miR-200b#	−17,569.40	9.2×10^{-27}	Chr. 1(p36.33)	37
miR-7-2#	−47,261.60	2.6×10^{-20}	Chr. 15(q26.1)	38
miR-767-3p	−94,315.40	2.2×10^{-39}	Chr. X(q28)	39

Finally, of the miRNAs presented in Table 2, miR-375 has been reported to be associated with a function in the pituitary gland [50], and it has been shown to regulate the expression of pro-opiomelanocortin (POMC), a common precursor of melanocortin-related peptides in the pituitary [50].

Comparisons of the microRNA expression profile between craniopharyngioma clustering groups and the controls

Unsupervised hierarchical clustering showed three clustering groups (Fig. 1). Pairwise comparisons were performed between each clustering group (1, 2, and 3) and controls to determine the differentially expressed miRNAs in each of the clustering groups. In Table 3, we present the ten miRNAs that are most differentially expressed when these comparisons were performed. The results indicate downregulation of miR-132 in both group 1 and group 2 tumors versus controls. It was also found to be downregulated when all craniopharyngiomas were compared to controls (Table 2). It is known that the early pituitary gland undergoes epithelial to mesenchymal transition (EMT) to form glandular tissue [51]. Interestingly, miR-132 has previously been shown to suppress EMT in pituitary tumors [52]. miR-132 has also been shown to be involved with colorectal cancer [53] and lung cancer [54]. Renjie and collaborators [52] showed that miR-132 targets the *SOX5* gene-inhibiting cell proliferation, invasion, and migration of two pituitary epithelial cell lines (GH3 and MMQ), one of which (GH3) is a tumor cell line. In colorectal and lung cancer studies [53, 54], it has been demonstrated that miR-132 targets *ZEB2*, a known EMT regulator [55, 56]. Because miR-132 was found to be significantly downregulated when group 1 and group 2 were compared to controls, but not when group 3 (the "lower aggressiveness" clustering group) is compared to controls, this might suggest that downregulation of miR-132 is an indicator of aggressiveness in craniopharyngiomas.

Author's personal copy

Table 3 List of the most differentially expressed microRNAs when comparing each group from the hierarchical clustering with controls

miRNA	Fold change	*p* value
A—Group 1 vs controls		
miR-1274b	51,536,654	7.1×10^{-21}
miR-1260	10,519,510	3.9×10^{-35}
miR-1274a	172,008	2.3×10^{-14}
miR-1183	162,031	2.8×10^{-26}
miR-663b	148,620	4.6×10^{-28}
miR-320b	109,472	3.7×10^{-29}
miR-375	−178,468	2.4×10^{-16}
miR-767-3p	−191,407	8.8×10^{-27}
miR-7-2-3p	−191,666	6.8×10^{-20}
miR-132	−327,186	1.1×10^{-16}
B—Group 2 vs controls		
miRNA	Fold change	*p* value
miR-596	60,920,269	4.8×10^{-29}
miR-1274b	51,441	4.4×10^{-12}
miR-767-3p	−17,580	5.5×10^{-19}
miR-7-5p	−19,510	1.3×10^{-10}
miR-7-2-3p	−37,617	2.8×10^{-19}
miR-383	−57,208	1.2×10^{-12}
miR-885-5p	−61,172	3.4×10^{-12}
miR-488	−62,039	1.9×10^{-22}
miR-485-3p	−71,103	6.2×10^{-19}
miR-132	−7,927,529	1.5×10^{-22}
C—Group 3 vs controls		
miRNA	Fold change	*p* value
miR-596	145,433	1.9×10^{-9}
miR-1274b	73,996	1.1×10^{-21}
miR-1260	9361	4.3×10^{-31}
miR-424-3p	−5020	2.7×10^{-23}
miR-1225-3p	−5661	2.7×10^{-14}
miR-1243	−10,092	1.0×10^{-20}
miR-542-5p	−10,481	9.4×10^{-15}
miR-7-2-3p	−24,411	1.8×10^{-13}
miR-200b-5p	−36,504	1.4×10^{-26}
miR-767-3p	−117,314	1.8×10^{-32}

Also of importance, these pairwise comparisons showed upregulation of miR-596, which is among the ten most differentially expressed miRNAs when group 2 and group 3 were compared to controls but not when group 1 was compared to normal controls (Table 3). Upregulation of miR-596 is not in accordance with the published literature, which suggests that miR-596 is a tumor suppressor gene in oral cancer. In that study, Endo et al. [57] found that a CpG island 5′ to the miR-596 gene was hypermethylated in oral squamous cell carcinoma cell lines and primary oral squamous cell carcinoma cases when compared to a normal counterpart cell line. Furthermore, the authors demonstrated that the ectopic expression of miR-596 dsRNA in vitro induced significant growth inhibition in the two cell lines compared to the control with increased apoptosis. The ectopic expression of the miR-596 dsRNA also resulted in the inhibition of tumor growth in vivo [57].

Conclusion

To date, this is the first paper that explores an extensive analysis of miRNAs in craniopharyngiomas. Downregulation of miR-132 does appear to be a marker of more aggressive craniopharyngiomas and also plays a role in epithelial–mesenchymal transition. It is known that the early pituitary gland undergoes EMT to form glandular tissue. As craniopharyngiomas are regarded as a developmental disease, we hypothesize that downregulation of miR-132 disrupts this normal process and at least in part contributes to the development of craniopharyngioma. Further studies investigating the role of miR-132 in pituitary cell lines might shed some light to the etiology of craniopharyngiomas and prediction of more aggressive behavior in craniopharyngiomas. In addition to the findings related to miR-132, there are several other dysregulated miRNAs in craniopharyngiomas when compared to controls. Further research needs to be performed to determine if there is a miRNA pattern that leads to the development of craniopharyngioma or function as markers of aggressiveness. This will help to determine if there are novel therapeutic options that can be explored in the treatment of craniopharyngiomas.

Acknowledgments The authors would like to acknowledge the significant contribution and dedication of Elio F. Vanin PhD who passed away right after completing the draft of this manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Funding The Matthew Larson Foundation; Dr. Ralph and Marian C. Falk Medical Research Trust and the Maeve McNicholas Memorial Foundation.

Ethical standards This study has been approved by the Ann and Robert H. Lurie Children's Hospital of Chicago Institutional Review Board and had therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki. All persons gave their informed consent prior to their inclusion in the study.

References

1. Karavitaki N, Cudlip S, Adams CB, Wass JA (2006) Craniopharyngiomas. Endocr Rev 27:371–397
2. Kelberman D, Rizzoti K, Lovell-Badge R, Robinson IC, Dattani MT (2009) Genetic regulation of pituitary gland development in human and mouse. Endocr Rev 30:790–829

Author's personal copy

3. Watkins-Chow DE, Camper SA (1998) How many homeobox genes does it take to make a pituitary gland? Trends Genet 14: 284–290
4. Wang Y, Liang Y, Lu Q (2008) MicroRNA epigenetic alterations: predicting biomarkers and therapeutic targets in human diseases. Clin Genet 74:307–315
5. Ambros V (2004) The functions of animal microRNAs. Nature 431:350–355
6. Jansson MD, Lund AH (2012) MicroRNA and cancer. Mol Oncol 6:590–610
7. Croce CM (2009) Causes and consequences of microRNA dysregulation in cancer. Nat Rev Genet 10:704–714
8. Costa FF, Bischof JM, Vanin EF, Lulla RR, Wang M, Sredni ST, Rajaram V, Bonaldo Mde F, Wang D, Goldman S, Tomita T, Soares MB (2011) Identification of microRNAs as potential prognostic markers in ependymoma. PLoS One 6, e25114
9. Sredni ST, Bonaldo Mde F, Costa FF, Huang CC, Hamm CA, Rajaram V, Tomita T, Goldman S, Bischof JM, Soares MB (2010) Upregulation of mir-221 and mir-222 in atypical teratoid/rhabdoid tumors: potential therapeutic targets. Childs Nerv Syst 26: 279–283
10. Sredni ST, Huang CC, Bonaldo Mde F, Tomita T (2011) MicroRNA expression profiling for molecular classification of pediatric brain tumors. Pediatr Blood Cancer 57:183–184
11. Campanini ML, Colli LM, Paixao BM, Cabral TP, Amaral FC, Machado HR, Neder LS, Saggioro F, Moreira AC, Antonini SR, de Castro M (2010) CTNNB1 gene mutations, pituitary transcription factors, and MicroRNA expression involvement in the pathogenesis of adamantinomatous craniopharyngiomas. Horm Cancer 1:187–196
12. Saeed AI, Bhagabati NK, Braisted JC, Liang W, Sharov V, Howe EA, Li J, Thiagarajan M, White JA, Quackenbush J (2006) TM4 microarray software suite. Methods Enzymol 411:134–193
13. Rittinger O, Kranzinger M, Jones R, Jones N (2003) Malignant astrocytoma arising 10 years after combined treatment of craniopharyngioma. J Pediatr Endocrinol Metab 16:97–101
14. Luu QT, Loredo LN, Archambeau JO, Yonemoto LT, Slater JM, Slater JD (2006) Fractionated proton radiation treatment for pediatric craniopharyngioma: preliminary report. Cancer J 12:155–159
15. Beckers A, Lodish MB, Trivellin G, Rostomyan L, Lee M, Faucz FR, Yuan B, Choong CS, Caberg JH, Verrua E, Naves LA, Cheetham TD, Young J, Lysy PA, Petrossians P, Cotterill A, Shah NS, Metzger D, Castermans E, Ambrosio MR, Villa C, Strebkova N, Mazerkina N, Gaillard S, Barra GB, Casulari LA, Neggers SJ, Salvatori R, Jaffrain-Rea ML, Zacharin M, Santamaria BL, Zacharieva S, Lim EM, Mantovani G, Zatelli MC, Collins MT, Bonneville JF, Quezado M, Chittiboina P, Oldfield EH, Bours V, Liu P, WdH W, Pellegata N, Lupski JR, Daly AF, Stratakis CA (2015) X-linked acrogigantism syndrome: clinical profile and therapeutic responses. Endocr Relat Cancer 22:353–367
16. Trivellin G, Daly AF, Faucz FR, Yuan B, Rostomyan L, Larco DO, Schernthaner-Reiter MH, Szarek E, Leal LF, Caberg JH, Castermans E, Villa C, Dimopoulos A, Chittiboina P, Xekouki P, Shah N, Metzger D, Lysy PA, Ferrante E, Strebkova N, Mazerkina N, Zatelli MC, Lodish M, Horvath A, de Alexandre RB, Manning AD, Levy I, Keil MF, Sierra Mde L, Palmeira L, Coppieters W, Georges M, Naves LA, Jamar M, Bours V, Wu TJ, Choong CS, Bertherat J, Chanson P, Kamenicky P, Farrell WE, Barlier A, Quezado M, Bjelobaba I, Stojilkovic SS, Wess J, Costanzi S, Liu P, Lupski JR, Beckers A, Stratakis CA (2014) Gigantism and acromegaly due to Xq26 microduplications and GPR101 mutation. N Engl J Med 371:2363–2374
17. Odenthal M, Hee J, Gockel I, Sisic L, Schmitz J, Stoecklein NH, Driemel C, Mohlendick B, Schmidt T, Knoefel WT, Lang H, Buttner R, Ott K, Vallbohmer D (2015) Serum microRNA profiles as prognostic/predictive markers in the multimodality therapy of locally advanced adenocarcinomas of the gastroesophageal junction. Int J Cancer 137:230–237
18. Della Vittoria Scarpati G, Falcetta F, Carlomagno C, Ubezio P, Marchini S, De Stefano A, Singh VK, D'Incalci M, De Placido S, Pepe S (2012) A specific miRNA signature correlates with complete pathological response to neoadjuvant chemoradiotherapy in locally advanced rectal cancer. Int J Radiat Oncol Biol Phys 83: 1113–1119
19. Zhang X, Cheng J, Fu L, Li Q (2016) Overexpression of tissue microRNA10b may help predict glioma prognosis. J Clin Neurosci
20. Mao Y, Wu S, Zhao R, Deng Q (2016) MiR-205 promotes proliferation, migration and invasion of nasopharyngeal carcinoma cells by activation of AKT signalling. J Int Med Res 44:231–240
21. Wang N, Tao L, Zhong H, Zhao S, Yu Y, Yu B, Chen X, Gao J, Wang R (2016) miR-135b inhibits tumour metastasis in prostate cancer by targeting STAT6. Oncol Lett 11:543–550
22. Mirghasemi A, Taheriazam A, Karbasy SH, Torkaman A, Shakeri M, Yahaghi E, Mokarizadeh A (2015) Down-regulation of miR-133a and miR-539 are associated with unfavorable prognosis in patients suffering from osteosarcoma. Cancer Cell Int 15:86
23. Ma R, Zhang G, Wang H, Lv H, Fang F, Kang X (2012) Downregulation of miR-544 in tissue, but not in serum, is a novel biomarker of malignant transformation in glioma. Oncol Lett 4: 1321–1324
24. Chen L, Zhang J, Feng Y, Li R, Sun X, Du W, Piao X, Wang H, Yang D, Sun Y, Li X, Jiang T, Kang C, Li Y, Jiang C (2012) MiR-410 regulates MET to influence the proliferation and invasion of glioma. Int J Biochem Cell Biol 44:1711–1717
25. Wang C, Liu P, Wu H, Cui P, Li Y, Liu Y, Liu Z, Gou S (2016) MicroRNA-323-3p inhibits cell invasion and metastasis in pancreatic ductal adenocarcinoma via direct suppression of SMAD2 and SMAD3. Oncotarget
26. Sun M, Huang F, Yu D, Zhang Y, Xu H, Zhang L, Li L, Dong L, Guo L, Wang S (2015) Autoregulatory loop between TGF-beta1/miR-411-5p/SPRY4 and MAPK pathway in rhabdomyosarcoma modulates proliferation and differentiation. Cell Death Dis 6, e1859
27. Liu H, Song Z, Liao D, Zhang T, Liu F, Zheng W, Luo K, Yang L (2015) miR-503 inhibits cell proliferation and invasion in glioma by targeting L1CAM. Int J Clin Exp Med 8:18441–18447
28. Ma N, Zhang W, Qiao C, Luo H, Zhang X, Liu D, Zang S, Zhang L, Bai J (2016) The tumor suppressive role of miRNA-509-5p by targeting FOXM1 in non-small cell lung cancer. Cell Physiol Biochem 38:1435–1446
29. Wu WJ, Shi J, Hu G, Yu X, Lu H, Yang ML, Liu B, Wu ZX (2015) Wnt/beta-catenin signaling inhibits FBXW7 expression by upregulation of microRNA-770 in hepatocellular carcinoma. Tumour Biol
30. Yao H, Liu X, Chen S, Xia W, Chen X (2015) Decreased expression of serum miR-424 correlates with poor prognosis of patients with hepatocellular carcinoma. Int J Clin Exp Pathol 8:14830–14835
31. Zhai Q, Zhou L, Zhao C, Wan J, Yu Z, Guo X, Qin J, Chen J, Lu R (2012) Identification of miR-508-3p and miR-509-3p that are associated with cell invasion and migration and involved in the apoptosis of renal cell carcinoma. Biochem Biophys Res Commun 419: 621–626
32. Wu SL, Fu X, Huang J, Jia TT, Zong FY, Mu SR, Zhu H, Yan Y, Qiu S, Wu Q, Yan W, Peng Y, Chen J, Hui J (2015) Genome-wide analysis of YB-1-RNA interactions reveals a novel role of YB-1 in miRNA processing in glioblastoma multiforme. Nucleic Acids Res 43:8516–8528
33. Sun J, Feng X, Gao S, Xiao Z (2015) microRNA-338-3p functions as a tumor suppressor in human nonsmallcell lung carcinoma and targets Ras-related protein 14. Mol Med Rep 11:1400–1406
34. Xu Y, He J, Wang Y, Zhu X, Pan Q, Xie Q, Sun F (2015) miR-889 promotes proliferation of esophageal squamous cell carcinomas through DAB2IP. FEBS Lett 589:1127–1135

Author's personal copy

35. Cai J, Zhao J, Zhang N, Xu X, Li R, Yi Y, Fang L, Zhang L, Li M, Wu J, Zhang H (2015) MicroRNA-542-3p suppresses tumor cell invasion via targeting AKT pathway in human astrocytoma. J Biol Chem 290:24678–24688
36. Tombol Z, Eder K, Kovacs A, Szabo PM, Kulka J, Liko I, Zalatnai A, Racz G, Toth M, Patocs A, Falus A, Racz K, Igaz P (2010) MicroRNA expression profiling in benign (sporadic and hereditary) and recurring adrenal pheochromocytomas. Mod Pathol 23:1583–1595
37. Wu WR, Sun H, Zhang R, Yu XH, Shi XD, Zhu MS, Zeng H, Yan LX, Xu LB, Liu C (2016) Methylation-associated silencing of miR-200b facilitates human hepatocellular carcinoma progression by directly targeting BMI1. Oncotarget
38. Zhao J, Wang K, Liao Z, Li Y, Yang H, Chen C, Zhou YA, Tao Y, Guo M, Ren T, Xu L (2015) Promoter mutation of tumor suppressor microRNA-7 is associated with poor prognosis of lung cancer. Mol Clin Oncol 3:1329–1336
39. Kreth S, Limbeck E, Hinske LC, Schutz SV, Thon N, Hoefig K, Egensperger R, Kreth FW (2013) In human glioblastomas transcript elongation by alternative polyadenylation and miRNA targeting is a potent mechanism of MGMT silencing. Acta Neuropathol 125: 671–681
40. Schopman NC, Heynen S, Haasnoot J, Berkhout B (2010) A miRNA-tRNA mix-up: tRNA origin of proposed miRNA. RNA Biol 7:573–576
41. Sand M, Skrygan M, Sand D, Georgas D, Gambichler T, Hahn SA, Altmeyer P, Bechara FG (2013) Comparative microarray analysis of microRNA expression profiles in primary cutaneous malignant melanoma, cutaneous malignant melanoma metastases, and benign melanocytic nevi. Cell Tissue Res 351:85–98
42. Burmester T, Weich B, Reinhardt S, Hankeln T (2000) A vertebrate globin expressed in the brain. Nature 407:520–523
43. Cazzoli R, Buttitta F, Di Nicola M, Malatesta S, Marchetti A, Rom WN, Pass HI (2013) microRNAs derived from circulating exosomes as noninvasive biomarkers for screening and diagnosing lung cancer. J Thoracic Oncol Off Publ Int Assoc Stud Lung Cancer 8:1156–1162
44. Althoff K, Lindner S, Odersky A, Mestdagh P, Beckers A, Karczewski S, Molenaar JJ, Bohrer A, Knauer S, Speleman F, Epple M, Kozlova D, Yoon S, Baek K, Vandesompele J, Eggert A, Schramm A, Schulte JH (2015) miR-542-3p exerts tumor suppressive functions in neuroblastoma by downregulating Survivin. Int J Cancer 136:1308–1320
45. Bray I, Tivnan A, Bryan K, Foley NH, Watters KM, Tracey L, Davidoff AM, Stallings RL (2011) MicroRNA-542-5p as a novel tumor suppressor in neuroblastoma. Cancer Lett 303:56–64
46. Xu L, Li M, Wang M, Yan D, Feng G, An G (2014) The expression of microRNA-375 in plasma and tissue is matched in human colorectal cancer. BMC Cancer 14:714
47. Yu X, Wu Y, Liu Y, Deng H, Shen Z, Xiao B, Guo J (2014) miR-21, miR-106b and miR-375 as novel potential biomarkers for laryngeal squamous cell carcinoma. Curr Pharm Biotechnol 15:503–508
48. Wu C, Li M, Hu C, Duan H (2014) Clinical significance of serum miR-223, miR-25 and miR-375 in patients with esophageal squamous cell carcinoma. Mol Biol Rep 41:1257–1266
49. Mayne GC, Hussey DJ, Watson DI (2013) MicroRNAs and esophageal cancer—implications for pathogenesis and therapy. Curr Pharm Des 19:1211–1226
50. Zhang N, Lin JK, Chen J, Liu XF, Liu JL, Luo HS, Li YQ, Cui S (2013) MicroRNA 375 mediates the signaling pathway of corticotropin-releasing factor (CRF) regulating pro-opiomelanocortin (POMC) expression by targeting mitogen-activated protein kinase 8. J Biol Chem 288:10361–10373
51. Melmed S (2015) Pituitary tumors. Endocrinol Metab Clin N Am 44:1–9
52. Renjie W, Haiqian L (2015) MiR-132, miR-15a and miR-16 synergistically inhibit pituitary tumor cell proliferation, invasion and migration by targeting Sox5. Cancer Lett 356:568–578
53. Zheng YB, Luo HP, Shi Q, Hao ZN, Ding Y, Wang QS, Li SB, Xiao GC, Tong SL (2014) miR-132 inhibits colorectal cancer invasion and metastasis via directly targeting ZEB2. World J Gastroenterol 20:6515–6522
54. You J, Li Y, Fang N, Liu B, Zu L, Chang R, Li X, Zhou Q (2014) miR-132 suppresses the migration and invasion of lung cancer cells via targeting the EMT regulator ZEB2. PLoS One 9, e91827
55. Diaz-Lopez A, Moreno-Bueno G, Cano A (2014) Role of microRNA in epithelial to mesenchymal transition and metastasis and clinical perspectives. Cancer Manag Res 6:205–216
56. Hao J, Zhang Y, Deng M, Ye R, Zhao S, Wang Y, Li J, Zhao Z (2014) MicroRNA control of epithelial-mesenchymal transition in cancer stem cells. Int J Cancer J Int du Cancer 135:1019–1027
57. Endo H, Muramatsu T, Furuta M, Uzawa N, Pimkhaokham A, Amagasa T, Inazawa J, Kozaki K (2013) Potential of tumor-suppressive miR-596 targeting LGALS3BP as a therapeutic agent in oral cancer. Carcinogenesis 34:560–569