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# Solid Variant of Papillary Thyroid Carcinoma in a 15-Year-Old Boy: Diagnostic Utility of Immunohistochemical Panel

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## Abstract:

We present case of 15-year-old boy having solid-trabecular histological type in low-differentiated variant of PTC. The interest in the described case is also determined by the fact that in the preoperative examination of the patient thyroid nodular goiter and Hürthle cell adenoma of indefinite atypia by FNA (Bethesda III) was fixed, no PTC-induced changes and family history and radiation exposure was absent. In fact, we had three clinical-morphological versions: 1. Hürthle cell adenoma; 2. Nodular goiter; 3. PTC solid-trabecular variant. Differential diagnosis between medullary and anaplastic carcinomas should to be made by using of histological and immunohistochemical methods. A multicomponent immunohistochemical study have shown the high activity of TTF1, Cyclin D1 and thyroglobulin in combination with the activity of E-CAD indicating on the solid-trabecular variants of PTC, the epithelial phenotype and the transcription activity of thyroid-specific proteins were preserved, which is significant. Coordinated activity of TTF1 and Cyclin D1 is manifested by tumor tissue growth and differentiation and this phenomenon explains the tumor spread isolated characters and no local metastasis, which is one of highlight of our case. Therefore, the presented case does not have any features of undifferentiated carcinoma.

*Keywords: Solid variant, 15-year-old boy, Immunohistochemical panel.*

## INTRODUCTION

Papillary Thyroid Carcinoma (PTC) is the most common tumor of thyroid gland, about 80% of all malignant formation of thyroid gland. Besides the most common follicular and tall cell variants, PTC is one of the following histological options from Solid Variant of PTC (SVPTC), which is rare about 1-3% of PTC options and at the same time is most common in children. This option is not well studied. SVPTC tumor parenchyma present solid-trabecular components and the neoplastic cells revealed hypercellular nests with PTC's cytological patterns. SVPTC is first described by Carcangiu et al. in 1985. Later Nikiforov Y. et al. discussed SVPTC high rate (37%) in children and young people as a result of Chernobyl's nuclear accident. SVPTC has a less good prognosis than classical papillary carcinoma including shorter survival rate, however, much better than a low differentiated and antaplastic carcinomas. Therefore, a morphological assessment of unclear histological criteria of this variant is important to avoid more aggressive diagnosis and recommended total thyroidectomy for recurrence prevent.

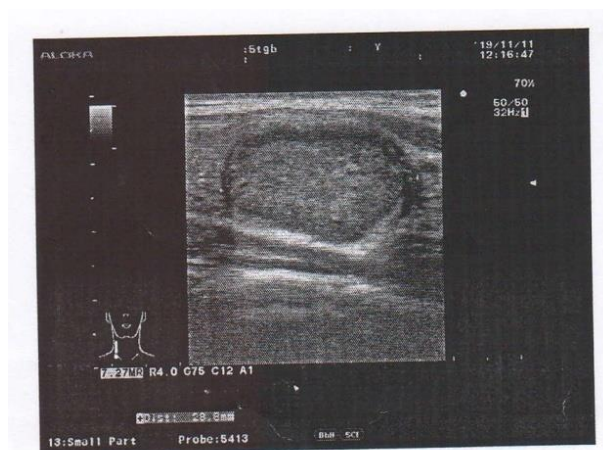
## Case Presentation

A 15-year-old boy was admitted for an anterior neck mass and discomfort during swallowing. He had no history of prior radiation to the head and neck and no known family medical history on endocrine disease. On physical examination, patient was medically stable with a blood pressure

of 125/60mmHg, pulse of 97 beats per minute and a temperature of 36.5°C. The thyroid gland moved upon swallowing. Thyroid gland of dense-elastic consistency. Left lobe – the nodule was well demarcated and measured about 3.0 cm in size. It was not adherent to any adjacent structures. Right lobe and isthmus appeared to be normal.

### Laboratory Results:

serum levels of free triiodothyronine, free thyroxine and thyrotropin, Vitamine D were within normal value range. Also, thyroglobulin and anti-thyroglobulin were negative. Ultrasound examination of the neck showed irregular hypoechoic nodule in the left lobe measuring 17X20X28 mm, with peri- and intranodular vascularization at colour-Doppler examination. The right thyroid lobe appeared normal. On the left side of the neck, in the middle third, behind the sternocleidomastoid muscle, there are two lymph nodes with a 7 mm length. Hilus slightly preserved and echogenic. From the left lobe of the thyroid gland caudally paratracheal lymph nodes were observed measuring 7 mm, 8 mm. and 7 mm. in size. Hilus slightly preserved and echogenic. From the right lobe of the thyroid gland caudally paratracheal lymph nodes were observed measuring 5 mm, 7 mm. and 7 mm in size. Hilus slightly preserved and echogenic (Fig. 1).



**Figure 1. Thyroid ultrasound image showing irregular hypodense lesion in the enlarged left lobe (Aloka SSD alfa 6-4D).**

Patient was screened for multiple endocrine neoplasia with negative results.

Thyroid aspiration (FNA) cytology documented lymphoplasmocytic infiltration, follicular cells hyperplasia, Hurthle cells metaplasia, atypia of undetermined significance by Bethesda Category III. Based on both cytopathological findings and ultrasound examination preoperative diagnosis was Nodular goiter Eo4.1 Patient underwent a total thyroidectomy, without neck dissection. Thyroid gland elastic consistency, in the lower pole of the large left lobe nodular formation up to 2 cm in size was detected. Right lobe in normal consistence.

### MATERIALS AND METHODS

Present case was reviewed and deemed exempt from written informed parents' consent by the Ethics committee and Board of medical sciences at Tbilisi State University based on Helsinki-Ethical Principles Declaration for Medical Research [7].

## Histological and Immunohistochemical Analyses

Gross examination showed a total thyroidectomy excised specimen weighed 88 g, with the right lobe measuring 45×20×30 mm, left lobe 60×30×20 mm and isthmus 30×20×11 mm.

Surgical specimens from left and right lobe and isthmus were fixed in 10% buffered formalin solution for 24 h and embedded in paraffin as routine. For histological examination 5-7 µm thick sections were stained with hematoxylin and eosin (H&E). Immunohistochemical (IHC) staining was performed on Formalin-fixed paraffin embedded (FFPE) tissue sections with antibodies against the following markers (Tab.1) Hematoxylin is used for nuclei counterstaining. Appropriate positive and negative controls were included for each reaction. All procedures were carried out in compliance with antibodies manufacturers' protocols.

**Table 1. Antibodies sources and manufacturers**

Primary antibody	Clone	Dilution	Source
Cytokeratin cocktail AE1/AE3	AE-1/AE-3	1:200	BioGenex, San Ramon, CA, USA
Chromogranin A	CGA/413+CHGA/777+CHGA/804	1:2000	BioGenex
Thyroglobulin	Polyclonal	1:8000	Dako, Denmark
Calcitonin	SKU: AN926GP	1:500	BioGenex
CEA	SKU: FG-365M-GP	1:20 000	BioGenex
S100	RTU-S100p	1:200	Biogenex
TTF1	SPT24	1:200	Leica, UK
Cyclin D1	P2D11F11	1:40	Leica, UK
CD56	CD564	1:50	Leica, UK
HBME-1	HBME-1	1:50	Dako, Denmark
Ki67	MIB-1	1:500	Dako, Denmark
E-CAD	NCH-38	1:200	Dako, Denmark

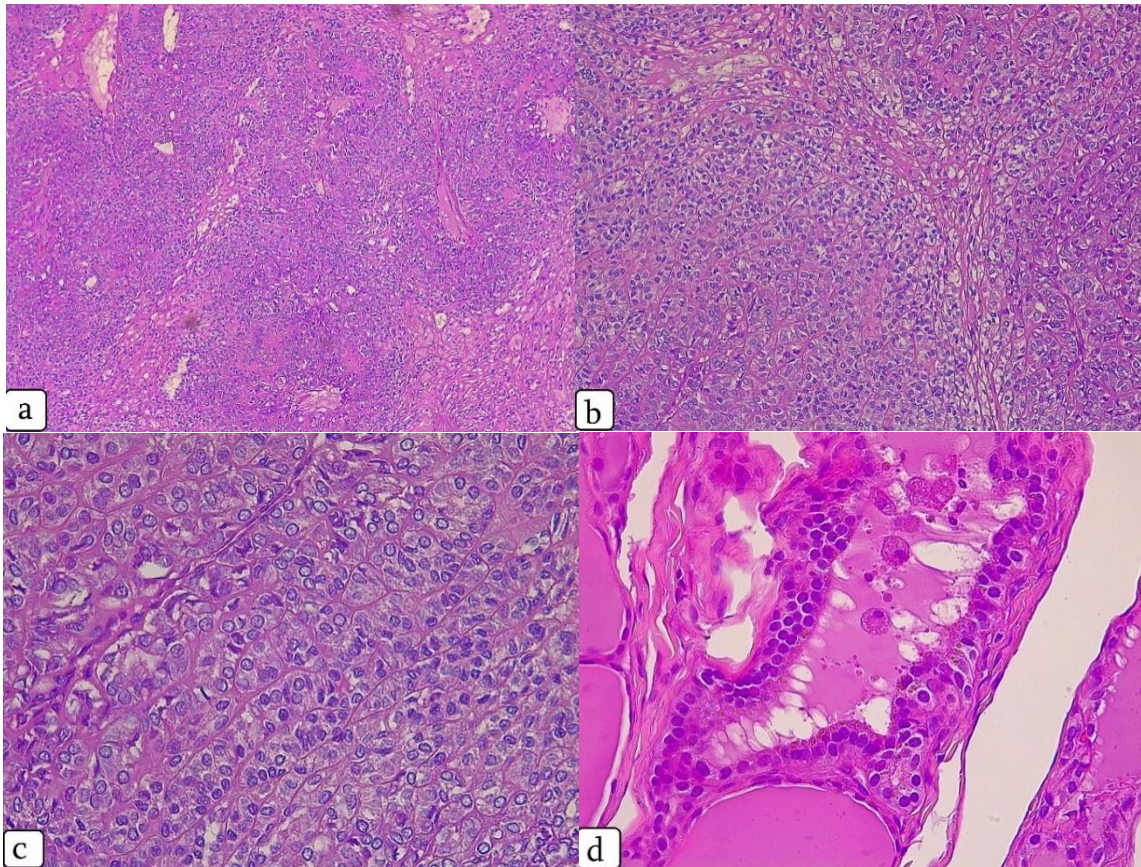
## Statistical Analysis

The intensity of staining was assessed semi-quantitatively by the evaluation of 150 cells in each high magnification field. The staining was regarded as positive in the case of nuclear/membrane expression. The results were expressed in – and + (-: no staining; -/+: staining present in 0–30% of cells, +: in 31–60% of cells, ++: in 61–90% of cells, and +++: in more than 90% of cells) (Figure 3).

## RESULTS AND DISCUSSION

Multiple tissue sections were taken for microscopic analysis. On histological examination: a nodule is seen in the left lobe surrounded by a fibrous capsule constructed of compactly arranged rounded and in some places with slightly elongated cells. The neoplastic follicular cells are in trabecular arranged or solid nests separated by fibrohyaline eosinophilic stroma. Tumor cells nuclei were large, optically clear, some irregular pleomorphic counterstain nuclei demonstrate grooves with distinct nucleolemma. Most of cells are located in perivascular area and initially reported as undifferentiated carcinoma with focal papillary area. Neither mitotic activity and nor area of necrosis are seen. The well-defined fibrous capsule was determined and separated neoplastic areas from the nonneoplastic parenchyma, which often present in PTC. It also showed foci of follicular cells hyperplasia, so-called stratification area (Figure. 2). Right lobe of the thyroid gland presents micro- and normofollicular structure.





**Figure 2. Solid variant of papillary thyroid carcinoma (SVPTC). H&E staining. a - The solid nests surrounded by fibrohyaline stroma; no papillary growth is present. b - The trabecular pattern of neoplastic cells. b, c – so-called ground-glass appearance of nuclei having classical nuclear features of PTC. d – Stratification area of follicular neoplastic cells with extensive fibrosis. a – X100; b – X200; c, d – X400.**

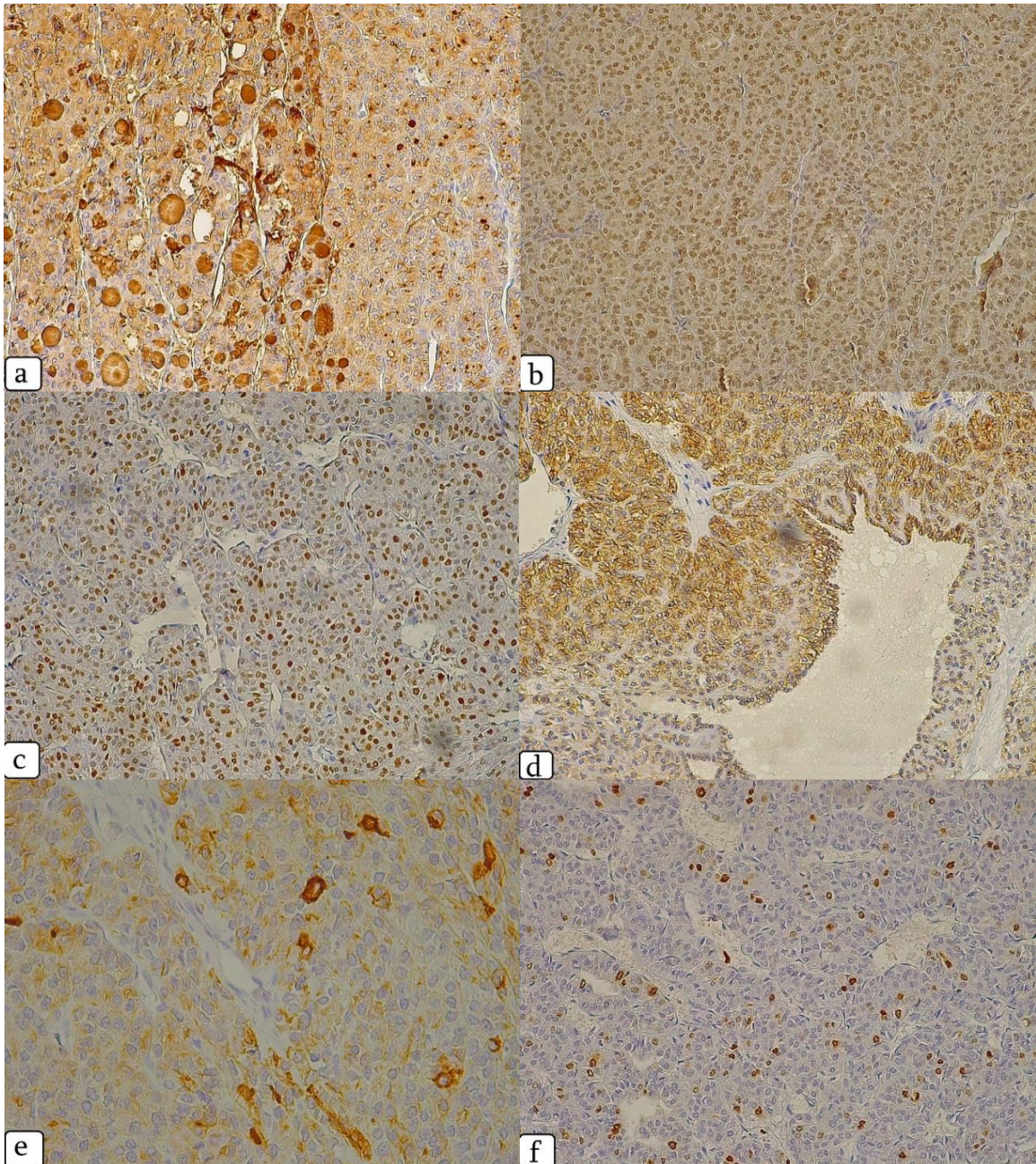
### Differential Diagnosis

Collision of nonclassical nuclear features of PTC, solid nest and trabecular arrangement of tumor cells, rarely reported undifferentiated carcinoma in children substantiated immunohistochemical examination in this case. Moreover, preoperative diagnosis was nodular goiter and FNA revealed Hurthle cells metaplasia and atypia of undetermined significance by Bethesda Category III.

### Immunohistochemical Study Results

Immunohistochemical staining showed the tumour cells were positive for Thyroglobulin (Tg), thyroid transcription factor-1 (TTF-1), Cyclin D1, E-CAD, Cytokeratin AE1/AE3, Ki-67 (Figure 3) and unequivocally negative expression of Calcitonin, Chromogranin A, S100 protein, HBME-1, CEA (carcinoembryonic antigen) (Figure 5). However, CD56 was positive in nonneoplastic and negative for neoplastic area, which is characterize for this antibody (Figure 5 d). Among the positive immunohistochemical results is noteworthy the strong cytoplasmic immunopositive reaction of thyroglobulin (Tg) in follicular cells, diffuse nuclear positivity of thyroid transcription factor 1 (TTF1) in SVPTC cells and diffuse positive expression of E-CAD in tumor cells, markedly reduced staining in the nonneoplastic area. High diffuse Cyclin D1 staining was in SVPTC cells nuclei. Focal immunopositive reactivity of cytokeratin AE1/AE3 was observed in the SVPTC trabecular area. As for the Ki-67 labeling index in tumor cells is very low ( $\approx 3\%$ ) (Figure 4).





**Figure 3.** The positive immunohistochemical results for SVPTC case. a – Strong cytoplasmic immunopositivity of Thyroglobulin (Tg) infollicular cells. b – Diffuse nuclear positivity of Thyroid transcription factor 1 (TTF1) in SVPTC cells. c – Diffuse higher Cyclin D1 expression in SVPTC cells nuclei. d – Diffuse positive expression of E-CAD in tumor cells, note the markedly reduce staining in nonneoplastic area. e – Focal immunopositivity of Cytokeratin AE1/AE3 in SVPTC trabecular area. f – The Ki-67 labeling index in tumor cells is very low (3%). Immunoperoxidase reaction. a-d, f – X200; e – X400.



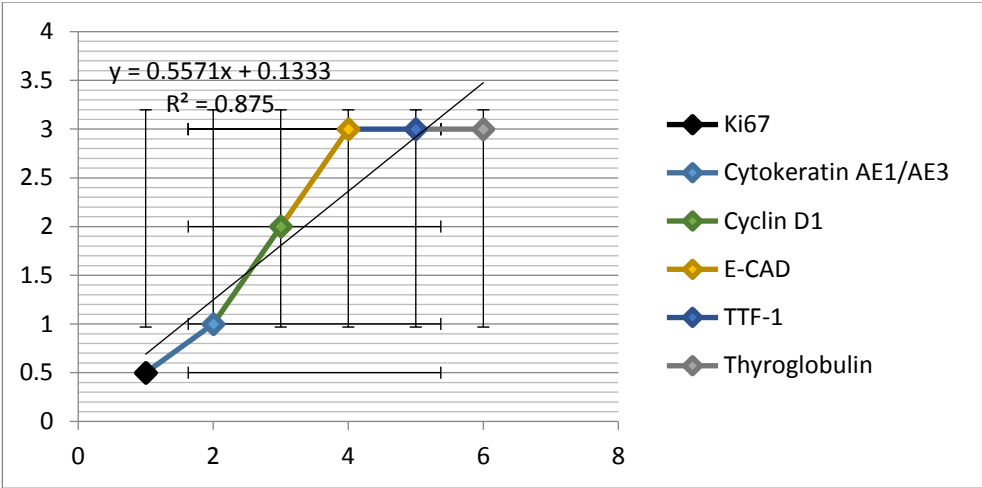
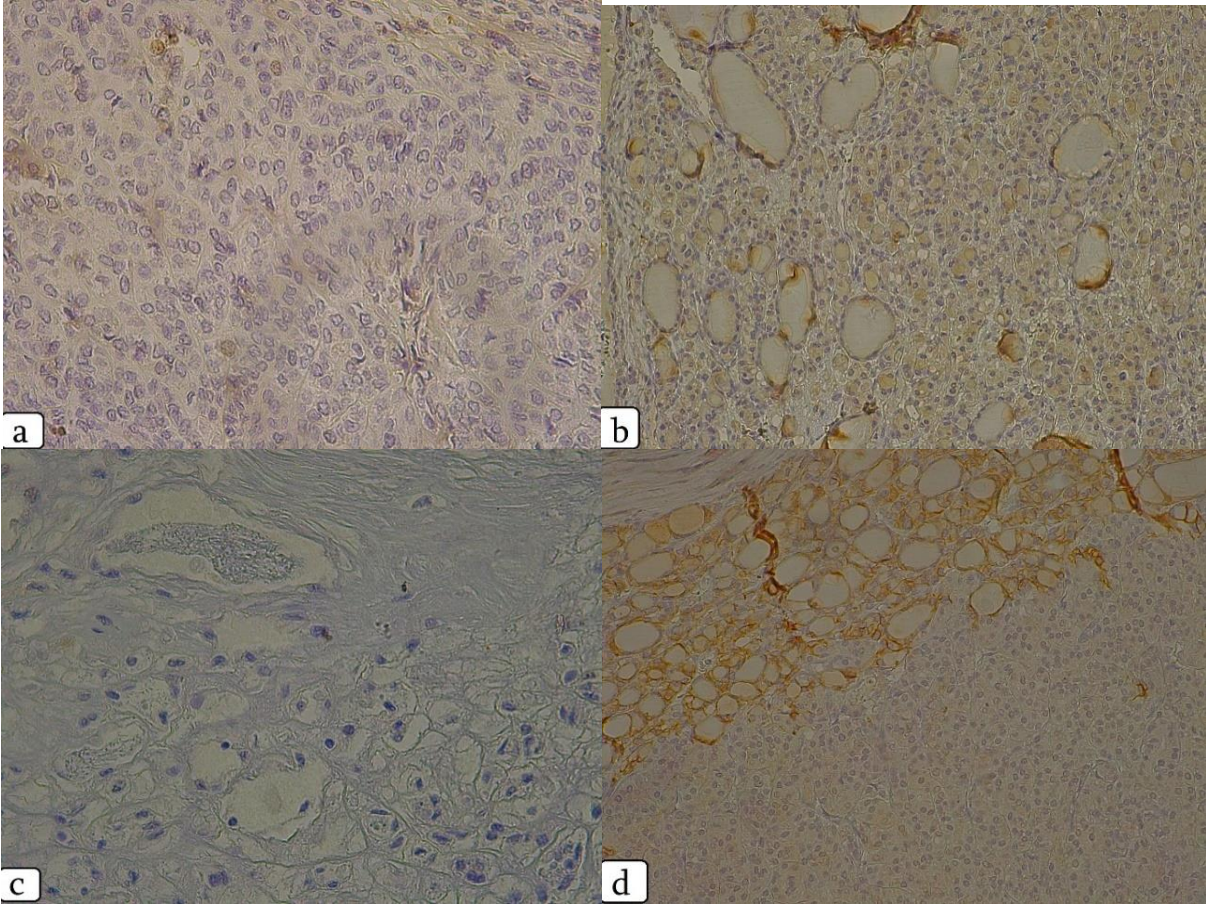


Figure 4. Immunostaining positive results for SVPTC case



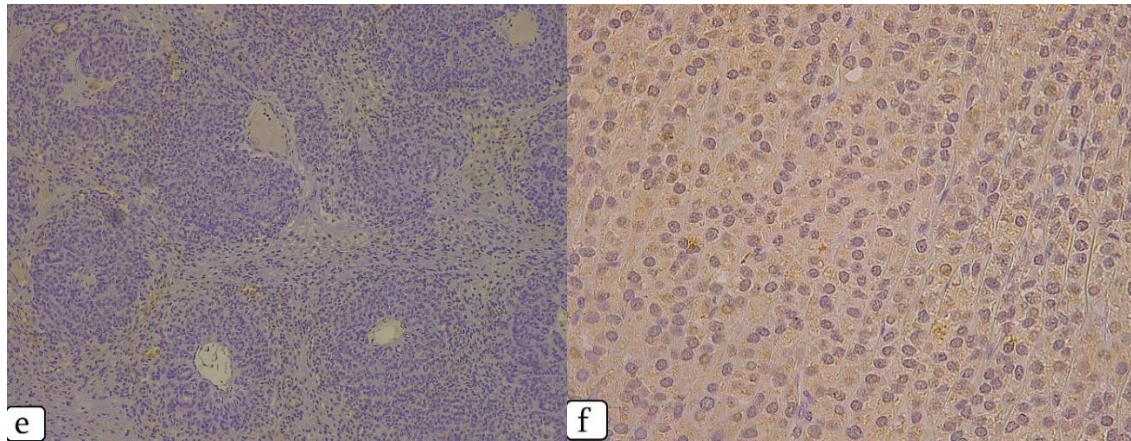


Figure 5. The negative immunohistochemical results for SVPTC case. a – Calcitonin. b – Chromogranin A. c – S100 protein. d – CD56 positive in nonneoplastic and negative for neoplastic area. e – Solid nest pattern negative for HBME-1. f – CEA negative immunoreactivity for tumor cells. Immunoperoxidase reaction. a, c, f - X400; b, d - X200; e – X100.

### Outcome and Follow-up

Postoperative radioiodine therapy and thyroid-stimulating hormone suppressive therapy were performed. During the follow-up period, the serum levels of thyroglobulin and anti-Tg Ab were routinely checked. Two months after the surgery patient was referred for a whole-body radioiodine scan and 48 h image after 2 mCi of radioiodine uptake. The child was treated with a dose of 80 mCi for remnant ablation and the post-therapy scan showed no additional iodine avoid lesions. Diagnostic I-131 scintigraphy (whole body) 2 months after surgery: Further diagnosis of iodotherapy. Reveals physiological involvement of the I-131 in the projection area of the oropharyngeal area, large salivary glands, digestive and urinary systems. In addition to physiological involvement, and a very weak area of I-131 accumulation is reflected centrally around the neck, corresponding to the post-thyroidectomy period (Fig. 6).

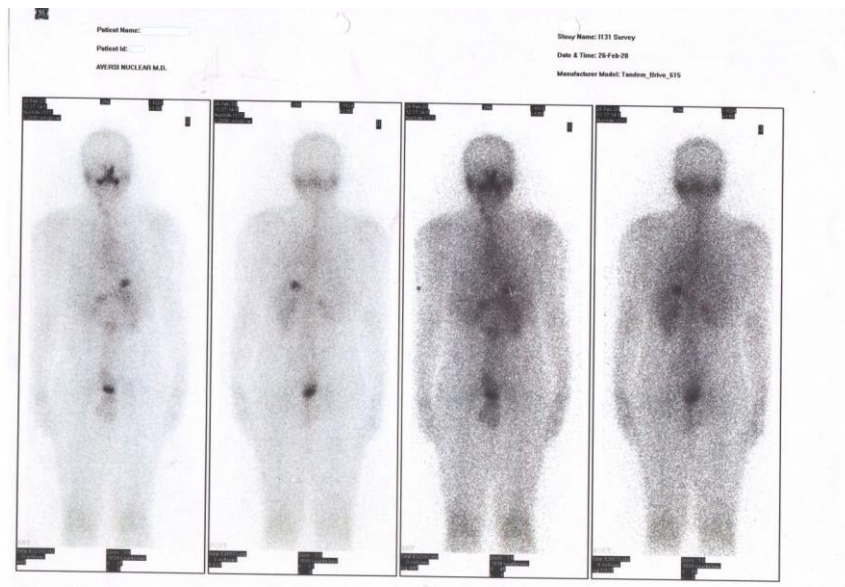


Figure 6. Diagnostic I-131 scintigraphy (whole body) 2 months after surgery 15-old-boy case  
Laboratory results 6 months after surgery: TSH – 2.25mU/L (0.35 - 4.0 mU/L); FT<sub>4</sub> – 1.04 ng/dL (0.89 - 1.76 ng/dL); Thyroglobulin - <0.20 ng/mL (<59.9 ng/dL); Anti-T.G – 11.45IU/mL (0 – 60.0 IU/mL); Vit. D – 27.39 ng/mL (30.0 – 100.0 ng/mL).



Patient is clinically well 12 months after surgery.

## DISCUSSION

We present case of 15-year-old boy having solid-trabecular histological type in low-differentiated variant of PTC. The interest in the described case is also determined by the fact that in the preoperative examination of the patient thyroid nodular goiter and Hürthle cell adenoma of indefinite atypia by FNA (Bethesda III) was fixed, no PTC-induced changes and family history and radiation exposure was absent. Intraoperatively unidentified tumor tissue was found; postoperative histological examination showed a PTC solid-trabecular variant of left thyroid lobe. In fact, we had three clinical-morphological versions: 1. Hürthle cell adenoma; 2. Nodular goiter; 3. PTC solid-trabecular variant (Figure 1). Extensive differential-diagnostic research has become necessary with low-differentiated carcinoma, medullary, anaplastic and metastatic tumors for identify both histological subtype as well as its molecular-biological profile. A multicomponent immunohistochemical study was performed, the follow results were obtained: TTF1, Thyroglobulin, E-CAD, Cyclin D1 have shown strong immunopositive reaction. Nuclear protein TTF1, as thyroperoxidase gene translation and thyroglobulin activator [8] shows a total sublimation with a high positive response in papillary carcinoma, in contrast to anaplastic and low-differentiated tumors [9-10]. In our case, E-CAD expression during PTC was preserved. It is noteworthy that, Batistatou et al, (2008) did not report a decrease in E-CAD expression during PTC [11]. It can be assumed, that E-CAD activity varies significantly between different subtypes of PTC, which can be a reason of high expression of the adhesive marker in certain aggressive forms, while in the clinically benign current case - sharply reduced, which is a similar view of other researchers [12]. The unequivocally high expression of Cyclin D1, presented in our case, indicates the activity of phosphorylation, transcription, and translation processes, including S phase and cell cycle progression. We can suggest that the increase in TTF1 translational activity is related to the synchronous reaction of Cyclin D1 and the coordinated activity of these two factors is manifested by tumor tissue growth and differentiation. This phenomenon explains the tumor spread isolated characters and no local metastasis, which is one of highlight of our case. The cytokeratin cocktail AE1/AE3 was also included in the differential-diagnostic panel, which showed weak (+) expression with ≈31–60% cell involvement. Use of this cocktail is limited if the aim of the study is not to confirm epithelial differentiation in the diagnostic panel. The low and high molecular weight clones so-called "pancytokeratin" involvement were excluded, which is critically important in the differential-diagnostic situation, although a low or negative reaction is still not enough to rule out the possibility of carcinoma. Low (3%) expression of Ki-67 is in full correlation with the above characteristics of the cell cycle (Figure 2 f). In our case, a negative reaction of the mesothelium marker HBME-1 was observed (Figure 4 e), thus limiting its use in aggressive forms of thyroid carcinoma.

## CONCLUSIONS

The solid-trabecular variant of PTC is relatively rare. Considering all the above-mentioned, differential diagnosis between medullary and anaplastic carcinomas should to be made by using of histological and immunohistochemical methods. Obviously, the high activity of TTF1, Cyclin D1 and thyroglobulin in combination with the activity of E-CAD indicates that in the solid-trabecular variants of PTC, the epithelial phenotype and the transcription activity of thyroid-specific proteins were preserved, which is significant. Therefore, the presented case does not have any features of undifferentiated carcinoma.

## Conflicts of Interest

The authors declare no conflict of interest.

## Acknowledgments

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# Role of Serotonin in Mammary Gland Development and Lactation Regulation in Dairy Animals: A Review

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## Abstract:

The aim of this review was to look in to the role of serotonin in the of mammary gland development and lactation regulation in dairy animals. In dairy animals, the dynamic interplay of endocrine hormones and locally produced factors such as serotonin regulates udder gland development and lactation. Serotonin (5-hydroxytryptamine, 5-HT) is a central neurotransmitter that influences mammary development and lactation in a variety of mammals. Its action is mediated by receptors found in a variety of mammary tissues. Serotonin played a significant role in the dilatation of the mammary alveolar space. Mammary 5-HT is used to regulate lactation, initiate the shift from the early stages of involution, and accelerate calcium mobilization from the skeleton. 5-HT is a nearby or local signal that controls milk secretion and promotes the formation of parathyroid hormone-related peptide (PTHrP) in several animals. According to various studies, serotonin influences milk yield and composition. Intrinsic 5-HT, which is generated inside the mammary epithelium, is vital in managing milk volume homeostasis in many species of mammals. Furthermore, when the udder gland is full of milk, 5-HT sends a negative feedback signal to the mammary epithelium, inhibiting further milk supply. Serotonin can limit milk production via a negative feedback loop by countering prolactin synthesis of mammary gland growth and lactation. Cellular connections (adherens junctions, desmosomes, and gap junctions), TPH1 induction, secretory activation, and pharmacological effects all have an impact on serotonergic control of lactation. Thus, serotonin linked with their receptors and play a crucial role in mammary gland development and lactation regulation.

*Keywords: Serotonin, mammary development, lactation*

## INTRODUCTION

Researchers set out to identify genes whose expression is influenced by prolactin in order to uncover possibly novel aspects of mammary gland physiology and development. Researchers focused their efforts on PRL-regulated genes that do not encode milk proteins or other milk-related gene products (Manabu et al., 2004). Serotonin (5-HT) is a peripheral (marginal) signaling chemical that impacts hemostasis, immune function, intestinal physiology, and other systems. 5-HT is a neurotransmitter that influences mood and cognition in both animals and humans (Hardman and Limbird, 2001). Because its receptors are found in a number of organs, including the endocrine, cardiovascular, immunological, and gastrointestinal systems, it is known as a marginal signaling molecule. Serotonin (5-HT) receptors are classified into four families (5-HTR<sub>1</sub>, 5-HTR<sub>2</sub>, 5-HTR<sub>3</sub>, 5-HTR<sub>4-7</sub>). Each serotonin receptor has its unique signaling pathways and physiological roles (Manabu et al., 2004). Serotonin (5-HT) is an old signaling chemical, although it is now frequently employed in mammary glands. The biological function of serotonin signaling and serotonin itself is complex, and extensive physiological, pharmacological, and molecular genetic studies on mammary process control have not been completed. For the first time, serotonin is extracted from the guts of several species, as well as their skins and serum. It was

named from two terms: serum-derived (sero-) vasoactive (tonic) chemical (Nelson and Robert, 2014).

The mammary gland's reactions to biosynthesis and secretion of serotonin are increased in response to alveolar space dilatation. Mammary serotonin has fulfilled two duties since its discovery some years ago; notably, it controls lactation and initiates the shift into the early phases of involution. Following that, serotonin stimulates the synthesis of parathyroid hormone-related peptide (PTHrP) via a local signal, causing the mammary gland to mobilize calcium from the skeleton (Nelson and Robert, 2014). Serotonin has a powerful role in both metabolism and physiology as a neurotransmitter, vasoconstrictor, gastrointestinal health regulator, and a mammary produced hormone that assists in lactation coordination (Horseman & Collier, 2014).

Furthermore, through performing on mammary epithelial cells (MECs), monoamine serotonin, a mammary-derived autocrine-paracrine signaling biomolecule, influence milk yield in many species. Bruschetta et al. (2020) discovered breed-specific serotonin concentrations in Italian Fresian and Brown Swiss cows during early lactation. Collier et al. (2012), in another cases, explored how serotonin ligands affect cow's milk output and composition. Serotonin also has a crucial metabolic function in the inhibition of the lactating mammary gland. Serotonin can also manage maternal metabolism without endangering maternal health (Hernández-Castellano et al., 2019). As a result, the main objective of this review paper was to examine the role of serotonin in mammary development and lactation regulation in lactating animals.

### SEROTONIN

Serotonin is an ancient signaling chemical found in a wide range of plants, animals, and unicellular creatures. Before 750 million years ago, the first serotonin receptor subtypes were found. Serotonin, a biogenic amine group, has affected the development and elasticity of many tissues in diverse animal species throughout the previous 2 billion years of development from protozoa to mammals. Serotonin is found in 5-hydroxytryptamine, Enteramine, Thrombocytin, 3-( $\beta$ Aminoethyl)-5-hydroxy indole, and Thrombotonin (Raymond et al., 2001).

Serotonin regulates a variety of physiological functions including homeostasis, nutrition, immunity, energy control, cardiovascular function, behavior, intestinal motility, and reproduction (Horseman and Collier, 2014; Wyler et al., 2017). The serotonin system's extra-neuronal role in the mammary gland functions was recently revealed. Serotonin is present in numerous organs' epithelial cells, including the mammary gland (Marshall et al., 2014). Intrinsic 5-HT is produced by the mammary epithelial cells of mice, cows, and humans. The intrinsic 5-HT generated by mammary epithelial cells is employed to regulate milk volume homeostasis. Since the mammary gland was full of milk, Serotonin suppressed milk production by delivering negative feedback to mammary epithelial cells (Lauder, 2004; Matsuda, 2004).

During lactation, fifty percent (50%) of the circulating serotonin is produced and released. Serotonins have a strong influence on mammary homeostasis inside mammary epithelial cells (Weaver et al., 2017). However, in non-lactating animals, the bulk of serotonin is produced and secreted by enterochromaffin cells in the intestine (Gershon & Tack, 2007).

#### The Role of Serotonin in Mammary Gland

Matsuda et al. (2004) identified an unexpected function of serotonin in mammary gland development Serotonin is a neurotransmitter produced locally by the mammary epithelium that



plays a key role in the negative feedback loop opposing prolactin stimulation of mammary gland growth and lactation. In response to gland fullness, this feedback loop suppresses milk excretion. Serotonin is widely recognized for its several activities, including the early development of oocytes and embryos in a wide range of animals, including sea urchins, starfish, mollusks, *Drosophila*, and rodents (Buznikov et al., 2001).

Similar to big animals, serotonin regulates preimplantation embryo development in mice (Vesela et al., 2003). In a range of animal species, including mice, cows, and humans, intrinsic serotonin (5-HT) generated within the mammary epithelium plays a significant physiological function in milk volume homeostasis. During lactation, the production of 5-HT (TPH1) increases due to the activation of tryptophan hydroxylase 1 by mammary epithelial cells. The TPH1 catalyzes the 5-HT production in the mammary gland. Serotonin is produced in mammary epithelial cells and transferred into the apical (milk) and basolateral spaces via the vesicular monoamine transporter. Suckling affects the quantity of serotonin in milk because serotonin levels in milk decrease during suckling (Chiba et al., 2018).

### Serotonergic Regulation of Lactation

#### ***Tryptophan Hydroxylase 1 (TPH1) Induction:***

The production of 5-HT in the mammary gland was identified using PRL-knockout (PRL-KO) mice (Matsuda et al., 2004). TPH1 was undetectable in PRL-knockout mice, but it was increased at the mRNA level when these mice were given large doses of PRL. TPH1 induction and 5-HT biosynthesis were both modulated by alveolar distension. TPH1 induction and 5-HT biosynthesis are triggered by mammary epithelial secretory activity. However, TPH1 induction and 5-HT biosynthesis are ultimately dependent on PRL. The period of time influences milk 5-HT secretion. The biggest rise in milk 5-HT occurred between 10 and 18 hours, when udder engorgement was at its maximum, as indicated in the figure below (Zia et al., 1987). As a result of the TPH1 regulatory pattern, 5-HT may play a role in the homeostatic control of milk secretion during milk stasis.

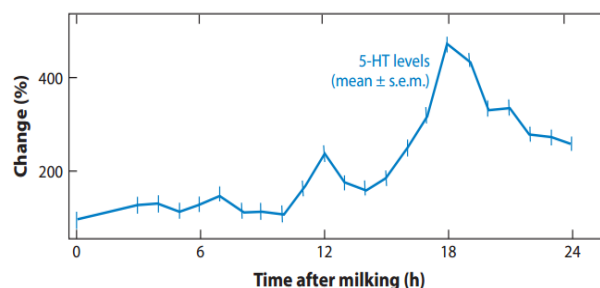


Figure 1: Milk serotonin (5-HT) during 24 h after milking (Zia et al., 1987)

#### ***Cellular Junctions:***

Cellular junctions preserve the membrane integrity between epithelial cells and the extracellular matrix. Because cellular connection features are critical in recognizing mammary epithelial physiology (Linzell and Peaker, 1971). Four junctions are involved in the serotonergic control of lactation. Adherens junctions (formerly known as intermediate junctions), desmosomes, gap junctions, and tight junctions are all types of junctions. These junctions were important in the control of serotonergic lactation. Adherens and desmosomes junctions join cells and give strength and rigidity to the epithelial membrane, whilst gap junctions enable substances to flow selectively between cells. Tight junctions also regulate the movement of molecules between the

luminal and serosal compartments by producing the z-axis barrier that divides the apical membrane from the basolateral membrane (Steed et al., 2010; Sawada, 2013).

During milk stasis, 5-HT dynamically modifies mammary epithelial tight junctions. Hernandez et al. (2011) employed FLX, an SSRI, to produce an in vivo tight junction response to 5-HT in milking cow udders. Serum lactose levels, a measure of tight junction leakiness, were significantly higher 24 hours after FLX infusion compared to values in mice given only the vehicle (Hernandez et al., 2011). Mammary epithelial cells were cultivated on permeable membranes to assess solute transport and transepithelial electric resistance. To validate the measurement, the Serotonin type-7 receptor (5-HT<sub>7</sub>) system was employed as a mediator of 5-HT tight junction responses. 5-HT agonists and antagonists can alter the basolateral membrane, but they have no effect on the apical membrane (Stull et al., 2007). Because of their strong constitutive activity as well as homologous and heterologous desensitization, 5-HT<sub>7</sub> receptors are employed as tight junction mediators (Pai et al., 2008; Krobert and Levy, 2002).

### ***Secretory Activation:***

Milk production is restricted during pregnancy due to placental steroid (particularly progesterone), which is part of endocrine signals (Freeman et al., 2000). However, no further study was conducted to determine specific local factors inhibit secretory activation. While research on mice mammary glands using pituitary grafting found that local serotonin, in concert with endocrine factors, can decrease mammary gland secretory activity when the gland is exposed to high amounts of lactogenic hormones (Matsuda et al., 2004).

### ***Drug Effects:***

Pharmacological drugs can be used to investigate mammary gland serotonin systems. Serotonergic drugs are commonly used as antidepressant medicines in therapeutic agents (Wong et al., 2005). They were commonly used to treat depression, anxiety, migraine headaches, fibromyalgia, and smoking cessation. These medications worked by decreasing the reuptake of released 5-HT, resulting in a longer-lasting 5-HT signal. In general, the concentration of Selective Serotonin Reuptake Inhibitors (SSRI) determines the drug effect on mammary gland serotonin, because low concentrations of SSRI increase tight junction resistance, whereas high concentrations of SSRI open a tight junction, resulting in decreased transepithelial resistance and tight junction protein disorganization (Marshall et al., 2010).

### ***Delayed Secretory Activation in New Mothers Taking SSRI:***

When a new mother is given an SSRI, the secretory stimulation of mammary gland serotonin into the udder might be delayed, which is contrary to laboratory findings. Lactation is delayed as a result of this occurrence. Because of the delayed lactation, mom/cow is at a considerable risk of failing to breast/udder feed their babies/calves (Marshall et al., 2010).

### ***Acceleration of Dry-off in the Dairy Cow:***

The obvious significance of 5-HT in natural lactation homeostasis and the effects of SSRI medication in new cows compelled researchers to investigate the system of serotonin mammary gland in dairy cows (Marshall et al., 2010; Hernandez et al., 2011). The cow, like humans and mice, expressed serotonin receptors (5-HT<sub>7</sub>) in its mammary epithelium. Cows, on the other hand, have an extra receptor that mice and humans do not have. When serotonin receptors (5-HT<sub>7</sub>) are activated, tight junctions in the cow mammary epithelium opened and milk protein gene production is suppressed. The secretory activation of the cow is delayed when it is given an SSRI.

In addition, the drying process in dairy cows is hastened. This means that the serotonin system, through the action of serotonergic drugs, might impact lactation performance of cow (Hernandez et al., 2011).

### Integration of Calcium Mobilization and Lactation Physiology by 5-HT

The physiological integration of tissue, cellular function, and lactation physiology at the organismal level can be modulated by mammary serotonin systems. Tryptophan hydroxylase 1 (TPH1) expression is induced by secretory material in the alveolar lumen and is controlled by lactational dilatation and contraction. Because of its unique mode of regulation, serotonin is a reliable source of information for the dynamic change that occurs during lactation within the mammary gland (Hernandez et al., 2012).

Serotonin, alveolar filling, and calcium mobilization are all connected to the induction of parathyroid-Hormone Related Peptide (PTHrP). During lactation, serotonin plays a significant role in PTHrP expression. As a result, its activity is directly mediated in epithelial cells via the 5-HT<sub>2B</sub> receptor type. PTHrP regulates serotonin's participation in whole-body calcium metabolism. The researchers begin by inducing PTHrP via serotonin signaling systems in order to solve the issue of why dairy cows are particularly sensitive to hypocalcemic crises whereas other animals are not (Horst et al., 2005). A big cisternal udder has been intentionally selected in dairy cows to drain the alveolar gaps efficiently. This reservoir must fill before the alveolar gaps may be completely distended. Inflating milk was the traditional therapy for milk fever a century ago. Figure 2 depicts, in a simplified form, the feedback loop through which milk secretion drives PTHrP.

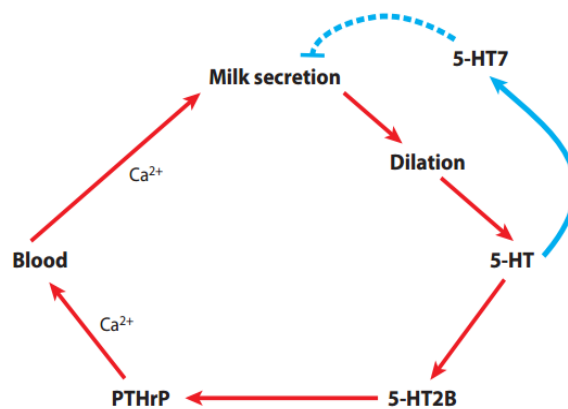


Figure 2. Linkage between serotonin mechanisms that regulate milk secretion via and 5-HT<sub>7</sub>

### CONCLUSIONS

This review was concluded by underlining the importance of serotonin in the development and lactation of dairy animals' mammary glands. In order to function on mammary tissues, serotonin is mediated via receptors located in a number of tissues. The intrinsic and mammary gland 5-HT signaling systems have paved the way for new study into the local regulation of physiology in lactating mammary glands, as well as communication between mammary glands. The 5-HT system in the mammary gland plays a significant role in the physiological integration of tissue and cellular activities, as well as lactation physiology at the organismal level.

Therapeutic 5-HT drugs can have an effect on lactation, either favorably or adversely. Pharmacological medications work by decreasing the reuptake of produced 5-HT in mammary

tissue, allowing for the generation of a longer-lasting 5-HT signal. As a result, for a long time, serotonin has been employed in mammary gland growth and milk yield advancement with few negative implications, as it prolonged the 5-HT signal to be generated by decreasing the rate of released 5-HT reuptake from mammary tissues.

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### Conflict of Interest

The author declares that there is no conflict of interest

### Ethics Approval

The article was written by a low-income author who requested a waiver for publication. Furthermore, the researcher's and the publisher's dignity, rights, safety, and well-being should be protected. However, because the work is a review article, the ethical standards for humans and animals were not taken into account.

### Consent to Participation

The author will take part in any actions required by the publisher regarding the article, as well as any other activities carried out by the publisher in line with the terms of the contract.

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### Availability of Data and Material (Data Transparency)

The data was transparent because the sources were properly recognized, yet the materials were not utilized.

### Code Availability (Software Application)

As per the review article, no software was used

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The author prepares this work entirely on his own, with no assistance from anyone

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# The Factual Demand and the Possibility of Self-Reliant in Cotton Cultivation in Bangladesh

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## Abstract:

The Bangladeshi garment is playing significant role in the country's development from the last decades. Today the garment sectors are providing 82% of the national export and count USD 30 billion revenues these numbers represent the sector as the most important manufacturing industry in Bangladesh. Cotton is the oldest and most important of the textile fibers. It is the back bone of the world's textile trade. Current survey shows that world production are about 25 million tones or 110 million bales annually, accounting for 2.5% of the world's arable land. Cotton is regarded as the golden arm for Bangladeshi textile because about 65% textile fabrics are made from cotton every day. Bangladesh requires 4-4.5 million bales (1 bale=217.7 kg or 480 pounds) of raw cotton which is slightly increasing. But Bangladesh is lack behind fulfilling the demand as Bangladesh are producing 1-2% of cotton requirement through the local production every year which is far behind the need. The remaining 98-99% requirement is fulfilled by the imported cotton mostly from Uzbekistan, India, USA, African countries, Turkmenistan, Australia etc. It is not possible to be self-reliant in raw cotton production as our raw cotton demand is very high and there is a land shortage for cotton cultivation but it can possible of production of initially 10 lac bales and finally 20 lac bales without hampering food production which is 50% of our national demand of raw cotton requirement and can save foreign currency of TK 11000 core per annum.

*Key words: Need, Demand, Arable, Area, Potential, CDB*

## INTRODUCTION

Cotton is like white gold for Bangladeshi textile. The numbers of factories are about five thousand of different sizes and about five million workers are working in these factories [1, 2]. These numbers show the most important fact that are playing the textile sector in the economy of Bangladesh. Bangladesh is on the verge of becoming developing country where textile playing the most significant role and cotton is the most important raw material of these textiles as 65% of products are made from cotton. Bangladesh needs about 4-4.5 million bales annually and most of these cottons are imported from other country. If importation is blocked due to any other reasons then the Bangladeshi garments will fall down. We have to keep that on mind. So we need to produce more cotton as possible. That's why we require cotton arable land to ensure cotton supply in danger when required to save our garments sector as well as our economy.

In this paper, we have tried to gather information about major cotton producer, major cotton consumer, demand of cotton and its condition in Bangladesh with price at the beginning section. The middle section of this paper will give us the importance of cultivation, prospects, production requirement, challenges of production and strategy to overcome. At the end of this paper we will have detailed concept about the growing behavior in divers' area, potential area of cotton



production in Bangladesh, climate change and mitigation strategy, cotton in income generation and the vision of Cotton Development Board (CDB)

## LITERATURE VIEW

### Cotton

The word "cotton" has come from the Arabian word. It is pronounced by the word "kutan", "qutn", "qutun" etc. in Arabian language. Cotton is a plummy soft natural staple fiber that grows in a boll or protective case around the seeds of the cotton plants of the genus *Gossypium* in the mallow family *malvaceae*. It is composed basically of a substance called cellulose [3]. Cotton gives food and fiber. Cotton also gives edible oil and seed byproducts for livestock food. The cotton seed can be crushed to extract vegetable oil and protein rich animal food. Cotton is grown - in tropical and subtropical regions which represent 2.5% of the all cultivated land. But among these the major producers of cotton are China, India, USA, Pakistan, Uzbekistan, Argentina etc.

### Kinds of Cotton

The plant is a shrub native to tropical and subtropical regions around the world. There are four basic types of cotton species are found that grows different part of the world. They are-

- *Gossypium hirsutum* is an upland cotton that grows well in Central America, Mexico, the Caribbean, and southern Florida, accounting for 90% of global output. The plant is generally 6 feet long and red in colour.
- *Gossypium barbadense*, often known as extra-long staple cotton, is endemic to tropical South America and accounts for 8% of global output. Plants range in length from 6 to 15 feet and are yellow in hue.
- *Gossypium arboretum*- known as tree cotton, grows well in India and Pakistan (representing less than 2% of global need).
- *Gossypium herbaceum*, often known as Levant cotton, is indigenous to southern Africa and the Arabian Peninsula (less than 2%). The plant's height ranges from 3 to 6 feet and its colour is yellow.
- Among these four varieties of cotton, two are grown in Bangladesh:
- *Gossypium hirsutum* and *Gossypium arboretum*, popularly known as comilla cotton.

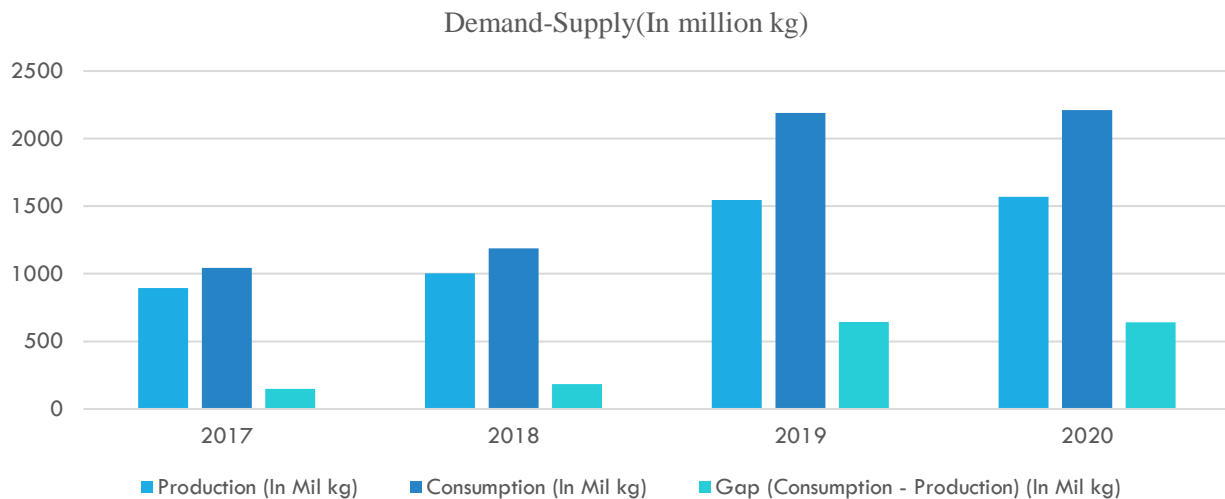
### The Demand of Cotton

The Garments and Apparel sector is the driving force of Bangladesh's economy as Bangladesh is the 2nd position among the world garments production country immediately after china [4, 5]. Most of this garments products are made from cotton fiber rather than other fiber. Bangladesh is in first position for its cotton importation among the other countries in the world. At present there are about 5000 thousand garments industry in Bangladesh where about 5 million people works to earn their livelihood. Here also 446 knit factories [6], 1343 weaving industry and about 3 lakh hand driven loom industry which need more numbers of yarn mostly cotton yarn. There are 425 spinning mills to ensure the supply of yarn for these industries [7]. By considering this above facts, the demand rate of cotton in Bangladesh is high in compare to other country. According to cotton development board, Bangladesh import 55 lakh bales cotton in the year of 2014 where this rate crosses in the year of 2016 when Bangladesh import 60 lakh bales cotton. In 2016-17 fiscal years, Bangladesh imported 1 thousand 361 metric tons cotton. The importance of cotton is increasing day by day. Vietnam is in the 2nd position for cotton importation as they imported 1 thousand 197 metric tons where china is in the 3rd position as they imported about 1 thousand 96 metric tons cotton. By seeing the import rate of cotton, it can easily be realized that cotton is much important

for Bangladeshi garments. H & M, Wal-Mart, Zara, Tesco, Next etc. well known garments merchandiser are related to these. Import of Raw Material for spinning are showed in Table-1 & Demand-supply situation of yarn in Bangladesh is represented in the below figure-1 [7].

**Table-1: A snap shot of import of raw materials for spinning in 2020**

Raw Material	Import (In Million Kg)	% of Total Import
Cotton Fiber	1401	89.0%
PSF Fiber	97	6.2%
Viscose Fiber	71	4.5%
Modal Tensile/Liocell Fiber	6	0.4%



**Fig.1- A snap shot of demand-supply situation of yarn in Bangladesh (In Million Kg)**

Now the textile industries are taking necessary steps to minimize the water consumption, industrial fuels usage, packaging elements and chemical usage rate as Accord alliance demand to establish themselves in the international level but no measures are taken to save the cotton wastage. At present Bangladesh are importing about 40% cotton from India and 20% are coming from Uzbekistan and West Africa. India is in the top by storing 5879 thousand metric tons cotton every year which are making them economically strong where the China and United State are in the 2<sup>nd</sup> and 3<sup>rd</sup> spot.

### Raw Cotton Condition in Bangladesh

The spinning mills use most of the imported cotton. In 2011-12 fiscal years Bangladesh produced 103 thousand bales and 2012-13 was 160 thousand bales. Our raw cotton demand is very high thus we can go for production initially 10 lac bales for short time and finally 20 lac tons for long term. This production is possible with the special help of government, policy makers, good research and extension management as well as development of marketing channel among various stakeholders like seed company, private ginner, farmer's input trader and so on. Good partnership between private public sectors needed to be improved. Southeastern zone, middle zone and northern part of Bangladesh are suitable for cotton production. The 33 districts of Bangladesh

mainly Kushtia, Chuadanga, Jhenaidah, Meherpur, Magura, Jessore, Rangpur, and Thakurgaon etc. are covering that area. The Chinese hybrid cotton is most mentioning factor for the growth of cotton on those districts.

### **Price of Cotton are Following Upward Trend**

Cotton price increased to 85.5 cents per pound, the highest in four years, mainly because of a looming trade war between the US and China, much to the vexation of Bangladesh's apparel makers. Cotton was traded between 83 cents and 84 cents a pound in the international markets. Cotton was traded between 70 cents and 71 cents a pound in November. Since the Chinese government in April announced potential 25 percent retaliatory tariffs on US goods, many in the cotton industry have wondered about the consequences, according to a report by the California Apparel News. If implemented, the tariffs would affect about \$50 billion in goods, \$16.5 billion of which includes crops and food items the US sends to China. Cotton price rose up in the global markets also because of volatile political situation in the world, currency fluctuation and stockpiling of the raw material by major global traders. "If the price spiral continues, Bangladeshi importers might face troubles as almost all the demand of the raw material is met through import in absence of domestic production. Local growers can meet only 3 percent of the local demand for cotton while 97 percent is imported mainly from India, the US, the Middle Eastern countries and some African countries. Annual cotton imports stand at more than \$3 billion. Bangladesh is the largest cotton importer in the world as China stopped importing the widely consumed white fiber. Bangladesh's cotton import will creep up to 7.1 million bales in 2017-18, further consolidating its position as the world's largest importer of the fiber, according to the United States Department of Agriculture [8].

### **The Importance of Cotton Cultivation**

Bangladesh aims to produce 1 million bales of cotton by the end of 2025, as the largest cotton importing country meets its total requirement from imports at present [9]. Currently, Bangladesh grows about 180,000 bales of cotton a year, which is just 1 percent of total demand in a year. Bangladesh spends more than \$3 billion to import cotton a year. A bale equals 480 pounds or 218 kilograms. Three major local groups -- Ispahani, Amber and Square began contract farming of cotton in different districts, mainly to minimize dependence on cotton imports. Many farmers in upland areas in different districts have already left tobacco leaf cultivation and are now growing cotton, which rakes in more profits for them, he. Of the annual import of cotton, majority percent comes from India as the neighboring country can supply quality cotton at affordable prices. "However, we must not rely on one or two sources as importers sometimes face challenges in case of any inconvenience in the supplying country. In order to reduce dependence on India, cotton imports from African countries increased significantly recently. Local spinners and traders import more than two million bales of cotton from African countries like Burkina Faso, Benin, Lesotho, Sudan and Chad. The import of cotton from the Commonwealth of Independent States like Uzbekistan will have to be stopped, as some major western garment retailers have embargoes on the use of cotton from Uzbekistan due to forced or child labor in cotton cultivation and harvesting. Australia and the US are also turning into big cotton sources for Bangladesh due to better crop quality. On future trends in cotton production and price, yield was good in different cotton producing countries; prices increased by 15 to 20 percent. There are 430 spinning mills at present in the country, which need more than 10 million bales of cotton. However, the spinning mills cannot run at full capacity due to an inadequate supply of energy and power. \$50 billion apparel export target by 2021 can be met if local spinners and weavers can supply yarn and fabric on time to the apparel makers. Cotton consumption in Bangladesh rose 4.91 percent year-on-year to 6.4

million bales in 2016 due to higher demand from spinners and garment makers, according to the US Department of Agriculture. Bangladesh has become the largest cotton importer in the world, as China stopped importing the fiber in recent years [10]. Bangladesh is fulfilling only 1% of our demand and to improve this to 10% Government is working collaborate with the Cotton Development Board. International Cotton Advisory committee presage that though production will increase but store rate of cotton will decrease in the universe this year. These premonitions encourage the trader to store the cotton in greater number for which the price increased. ICAC recently published cotton available and demand related premonition for the year 2017 to 2018 which show there will be decrease in the store of cotton for the 3rd time. This time the store will be 1 core 71 lakh 30 thousand tons which is least in number compare to the last 6 year. But there is a happy news that production will increase in the next season where the production will be 5% greater than now covering the 3 core 6 lakh hector area for cultivation. The ICAC optimist that the cultivation rate will increase in 7% in India includes 1 core 12 lakh hector area. The Indian farmer encourage in the production for increasing the cotton price. Beside India the production also increase in the USA in 2017-18 which includes 4% more than earlier as the production will 40 lakh tones. This time there is a possibility to cultivate cotton in 42 lakh hector land which will 10% greater than this season. In china cotton store rate will decrease including 1 core 71 lakh 30 thousand tons which will 50 lakh tones less than highest storage. Cotton crop not only provide fiber for the textile but also plays a role in the feed and oil industries with its seed, rich in oil (18-24%) and protein(20-40%). An estimated a large number of people are engaged in cotton production either on farm or in transportation, ginning, baling and storage.

### Prospects of Cotton Cultivation

1. Huge domestic requirements of cotton
2. Production per unit area is higher than the world average
3. Good quality fiber should be produced
4. Profitable than other crops
5. Cash crops
6. Contribute in food security
7. Contribute in employment generation and income generation
8. Profit can get at a time
9. Improve soil fertility, adaptive to climate change,
10. Saline and drought tolerant and multipurpose use;
11. Edible oil, oil cake, particle board etc. [11].

### Cotton Production Requirement

Cotton cultivation requires a long frost-free period, enough sunshine, and a moderate rainfall from 60 to 120cm. cultivation up to 1000m from sea level. Temperature for germination of seed is 16 to 30 degree with a minimum of 14 degree and maximum of 40 degree Celsius. For early vegetative growth, temperature must exceed 20 degree with 30 degree Celsius desirable. For proper bud formation and flowering the day time temperature should be higher than 20 degree and night temperature should be higher than 12 degree but should not exceed 40-27 degree respectively. Temperatures between 27 to 32 degree are optimum for boll development but above 38 degree are not expectable. Continuous or heavy rainfall and strong or cold winds seriously affect the cotton growth. Soils usually need to be fairly heavy although the level of nutrients doesn't need to be exceptional. Soil should be water holding and has retention toward moisture. Medium loams to sandy loam fertile soil are best for cotton production. Acid or dense sub soils

limit root penetration. The P<sup>H</sup> range is 5.5 to 8 The crop tolerant to soil salinity. The plant propagation by seed [12]

### **Challenges of Cotton Production**

Bangladesh prevail favorable weather for cotton production. Though 2.42 lakh hectares of land are suitable for cotton cultivation in Bangladesh 1 lakh hectares of land are available for cotton cultivation but cotton production are not increasing considerably due to many constraints related to research, marketing & management. The following are the major problems of cotton production in Bangladesh-

#### ***Scarcity of Land:***

The lands suitable for growing cotton have been declining due to urbanization, industrialization, housing and other purposes. Also farmers are intended to grow three or more crops from the same land in a year. But due to long duration cotton can't be fixed in the existing cropping pattern.

***Competition With Other High Value Crop:*** Cotton is highly competitive with other high value crops like- vegetables, spices, flowers, banana & other fruits. Farmers are migrated from cotton to these high value crops.

#### ***Lack of Short Duration, High Yielding & Pest Tolerant Varieties:***

CDB released 10 varieties for American Upland Cotton & 2 varieties for Hill Cotton. Out of these varieties, the farmers in Bangladesh cultivate only five varieties. Only CB-5 & CB-9 are hairy varieties which are slightly tolerant to sucking pests like jassids. None of the existing varieties are resistant to bollworm. Duration of all the varieties are about 6-7 months & low yield capacity compared with other countries.

#### ***High Input Cost:***

High input cost, particularly fertilizer and pesticides. This is because of the heavy reliance on pesticides for cotton pest management.

#### ***Insect Pests and Diseases of Cotton:***

The major insect pests of cotton in Bangladesh include the chewing insect namely American bollworm & spotted bollworm and the sucking insects namely the Jassids, Aphids & Whitefly. Farmers have to give more attention to the management of these pests & spent about 40% of the total production cost.

#### ***Weakness in Cotton Research:***

Adequate infrastructure & human resource facilities for cotton research is very limited within the Cotton Development Board. Most of the Scientist working under Cotton Development Board are junior in service and don't have much experience in research. Linkage between other Research Organizations is very low & inadequate facilities for human resource development [13].

### **Is Cotton Production Viable for Bangladesh while we have Limited Land?**

First of all, the yield of cotton in Bangladesh has improved significantly in recent years from 5-6 Mon per Bigha to 15-18 Mon per Bigha with the good work of Cotton Development Board and active leadership of current Executive Director.

This effort of improved yield needs to be carried out in future to double the productivity to 30 Mon per Bigha (Say; 25 and 30 Mon per Bigha during 2022-2030 and 2031-2041, respectively) from the current level that would add to the long term viability of cotton farming in Bangladesh.

As per USDA January 2022 report 1 hector of land produced 2,217 kg, 1,976 kg, 1,804 kg and 1,720 kg of cotton in Australia, China, Turkey and Brazil, respectively, in 2020/21 while Bangladesh can produce only 900 kg of cotton which is better than many other countries like; India (462 kg), Pakistan (445 kg), etc.

The quality of our cotton is good and very much suitable for ring yarn where our average count is Ne 30/s and major count range of Ne 6/s-12/s, Ne 20/s-40/s and Ne 16/s-40/s for denim, knit and woven yarn.

Secondly, in order to secure raw material for the export oriented spinning mills, having domestic production of 24% (2.4 million bales of cotton production per year) is not a bad option if it is viable. Major spinning countries are having their own raw material; i.e. China, India, Pakistan, Turkey, except Bangladesh and Vietnam.

Thirdly, if we don't focus on domestic cotton production, we would be spending roughly USD 4 billion (8-10 million bales) annually to import cotton.

If we can substitute 24% of our cotton use with domestic production by allocating total 11 lac Bigha land for only six months for cotton cultivation (Say; 5.5 and 11 lac Bigha, respectively during 2022-2030 and 2031-2041, respectively) to produce 2.4 million bales of cotton, we can save USD 1.27 billion at the same time add the same amount in Gross Domestic Product (GDP).

In addition, we would get edible oil, cottonseed oil cake, firewood, fertilizer, and create employment and business opportunity in the rural areas.

Fourthly, Global cotton production and consumption is confined within 120-126 million bales (Around 127,000 MT). So, fundamentally cotton is limited while 89% of raw material of our spinning mills is cotton.

Fifthly, considering per capita domestic fiber consumption @ 2 kg local consumption of cotton for 18 crore people is 1.6 million bales per year.

### **Strategies to Face the Challenge**

Cotton remains the chief raw material for textile industry. The demand for cotton textile products is increasing every day due to increasing global population. To meet the challenges ahead, viable strategies are need to be designed. These can be classified under the following heads.

- (a) Net productivity increase
- (b) Quality improvement
- (c) Resistance to biotic and abiotic stresses
- (d) Utilization of genetic engineering procedures

### ***Net Productivity Increase:***

The productivity increase sought may be in terms of seed cotton yield or lint yield. Manipulation of boll number, boll weight and sympodial branches provide opportunities of increasing

productivity. In cotton, source is not a problem but sink is a constraint. Genetic variability for boll retention has to be exploited along with balance between boll number and boll weight. Fiber yield increase can be achieved through increasing ginning out turn. The possibilities of upgrading ginning out turn up to 39 per cent in *G. hirsutum* has been demonstrated by varieties like Sahara in South Zone.

#### **Quality Improvement:**

Quality in terms of fiber properties and seed cotton oil are gaining importance. Innovations in spinning technology, such as open-end spinning, jet spinning etc., require higher levels of fiber strength. Hence, high priority to breeding cotton for increased strength and maturity assumes importance. The potential of improving fiber properties especially length and fineness, of diploid cultivated cotton needs to be exploited. The presence of gossypol in the oil is a negative attribute in terms of edible quality. The processing cost to remove gossypol from oil can be eliminated if the seeds have no gossypol glands. But gossypol is the main trapezoid aldehyde which imparts resistance against insects.

#### **Abiotic Stresses:**

Drought stress affects crop in rain fed areas and salinity in irrigated areas. Hence, resistance sources for both environments have to be utilized appropriately. Diploid cultivated cottons, which have stress tolerance capacities, occupy a sizable (27 lakh ha) area under rainfed conditions. Genetic improvement of these diploids (*G. arboreum* and *G. herbaceum*) needs to be given high and urgent priority so that these important genetic resources are not lost. Resistance breeding for salinity is as yet an untouched area. Under the biotic stresses, insects, like bollworms and diseases like CLCuV need urgent consideration for sustainable cotton production. Resistance to whitefly, which is the vector of CLCuV, provides solution to two problems simultaneously. Search for characters in wild *taxa* is ideal for breeding against biotic stresses. Transfer of palisade layer from *G. arboreum* to *G. hirsutum* is the best option for developing resistance to sucking pests.

#### **Utilizing Genetic Engineering Procedures:**

Though *Bt* cottons are already in the field, they are associated with threats of insecticide resistance development against bollworms and instability in their bio efficacy. Hence, use of alternate sources of insecticidal proteins like *tea* complexes of *P. luminacence* and *multiple gene constructs* (of Cry genes) can be thought of, to overcome the negative points of *Bt* cotton technology. Sucking pests may also be brought under the umbrella of biotechnology, to build resistance in cotton genotypes. Exploitation of *chitinase* gene against diseases is another good option for breeding fungal disease resistance. Antisense technology can be employed to construct varieties resistant to leaf CLCuV disease.

#### **Miscellaneous:**

To achieve "Total Natural Fiber" concept, breeding naturally colored cotton deserves consideration. Increasing spin ability of the colored cottons in addition to enhancing their productivity and range of colors are good areas for future work. Cotton is mostly cultivated as sole crop but area under cotton-based cropping system is also large. Development of genotypes suitable for different cropping system under different soil conditions deserves attention for increasing the net income per unit area. The concept of ultra-narrow spacing is the talk of the day. Short compact genotypes amenable for machine picking are important. These genotypes not only reduce dependence on labor for cotton picking but also fit in various cropping systems.

Accelerated efforts have to be made for the overall improvement of cotton to reach at least the world average of productivity [13].

### **Growing Behavior of Cotton in Diverse Area**

Thrust areas for cotton farming are needed to be analyzed and allocated 5.5 and 11 lac Bigha of land during 2022-2030 and 2031-2041, respectively like;

#### ***Drought Area:***

Cotton is considered as drought tolerant crop as the plant requires very minimum water as it has vertical tap root that provides resilience against drought and it has 'Compensatory growth'. Cotton responds to loss of fruiting parts (buds, flowers, bolls). Based on those characters cotton could be successfully grown in drought area. As we know rice is considered as highly water consuming crop and it requires 3000 liters of water to produce 1 kg rice. The farmers of those area get yield of 5-6 maunds per bigha which is not profitable for them. But to grow cotton it requires only one or two irrigation for the whole duration of crop. Last couple of years CDB has conducted many adaptive trials in Godhagari, Porsha and Neyamatpurupazilla to see the performance of cotton production in those areas. Farmers have gotten maunds per bigha which is highly profitable for the farmers of that area. Under high barind area there is two lac ha of land which is suitable for growing cotton. From that area at least 50000 ha land we want to take under cotton production [14].

#### ***Cotton in Hill Area:***

Cotton has been growing in three hill districts since time immemorial with jhum cultivation in the hill slope. Jhum is an old age method of mixed cropping where 10-12 crops have been cultivating in hill slope and yield is very low due to competition of many crops and varieties of all crops including cotton are indigenous. The cotton species which is grown in jhum is non-spinning, short staple length and low market price. As it is grown in jhum with many crops, it gives very low yield and less economic benefit. Several adaptive trial conducted by CDB in the hill slope, rice cotton intercropping an alternative option has developed which provide more yield and income for the hill farmers. There are about 550000 ha of hill slope which is suitable for rice cotton intercropping. There is some plain land between two hills and beside the hill which is called hill valley is now under tobacco cultivation. These types of plain lands are found Dighinala and Panchari of Khagrachari district and Lama and Alikadam of Bandarban districts. The tobacco could be replaced by cotton in the hill valley. There is also an area 52000 ha which could be taken under cotton cultivation and increase the income of tribal farmers of three hill districts [15].

#### ***Replacement of Tobacco:***

Tobacco cultivation has negative impact on soil health which kills the soil microorganism and it is also create health hazard for the farmers but farmers grow tobacco as they get some special incentives from the tobacco company but they want to replace tobacco if they get the suitable crop which is profitable for them. Cotton is one of the important alternative options for tobacco replacement. There are 70 thousand ha of land in different area like Kushtia, Meherpur, Rangpur etc. which is under tobacco cultivation and gradually we can replace this tobacco area under cotton cultivation. The replacement of tobacco has already started in Daulatpur of Kushtia and Rangpur, Thakurgao as well as in three hill districts [15].

#### ***Cotton in Saline Area:***

Cotton is also considered as 2nd saline tolerant crop after barley among all crops. Many countries of the world cotton are cultivated cotton in saline area where other crops are not grown. Only



seedling stage of cotton plant is sensitive to salinity but other stage of the crop is not sensitive to salinity. The salinity level of up to 11 ds /me has no effect on yield and after that yield will be affected. So, in the saline area of Bangladesh easily we could take under cotton cultivation. In Bangladesh there are 5000 ha of land under saline area from that area we want to take 2000 ha under cotton cultivation Cotton in Char area Cotton is a deep rooted crop and uptake nutrients from subsoil and adds biomass to the top soil by their big leaves and improves the soil fertility. The char area where the land is not inundation in the month of July December-that chars easily could be taken under cotton cultivation. Last couple of years we have been conducting research and development in the char area and successfully cotton has grown with reasonable yield. In Bangladesh there are 100,000 ha of land under char area from where we could take 20000 ha of land for cotton cultivation [15]

**Cotton in Agro-Forestry:**

Cotton is grown in high land where water stagnation is not occurred. This land is also suitable for fruit orchard like mango, litchi, jujube and mehoghoni wood lot. At the beginning of the establishment of orchard there is a big space between the plants where easily cotton could be successfully grown with good yield. Last couple of years we did many experiments in different orchard and found successful production of cotton.

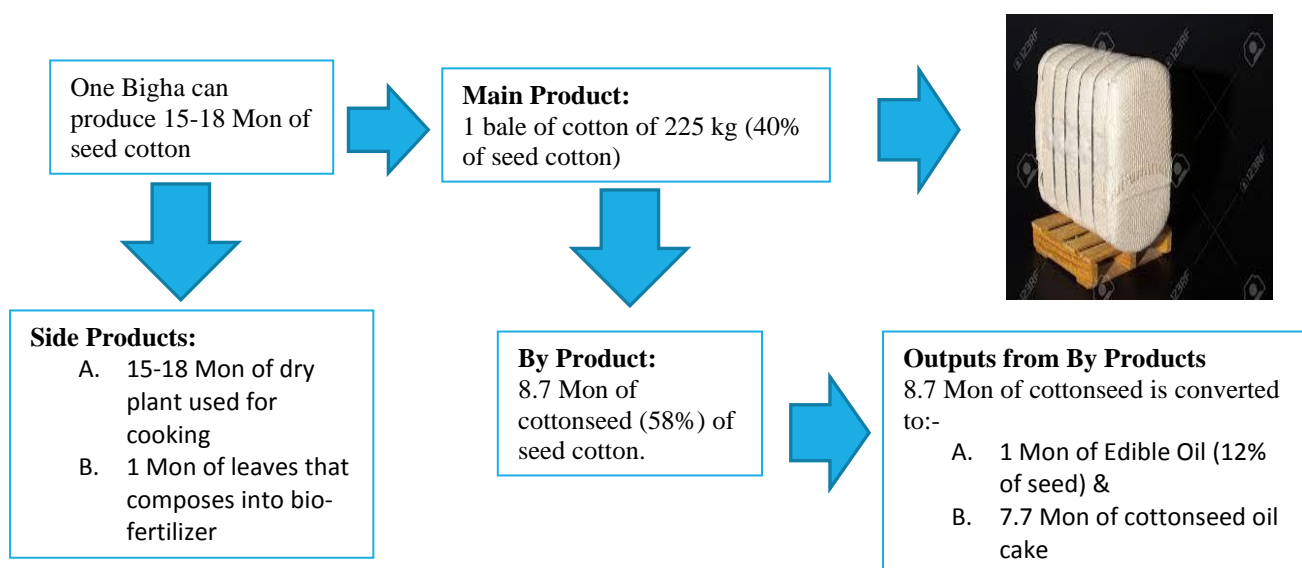


Fig.-2: Yield of seed cotton and conversion to raw cotton in bale

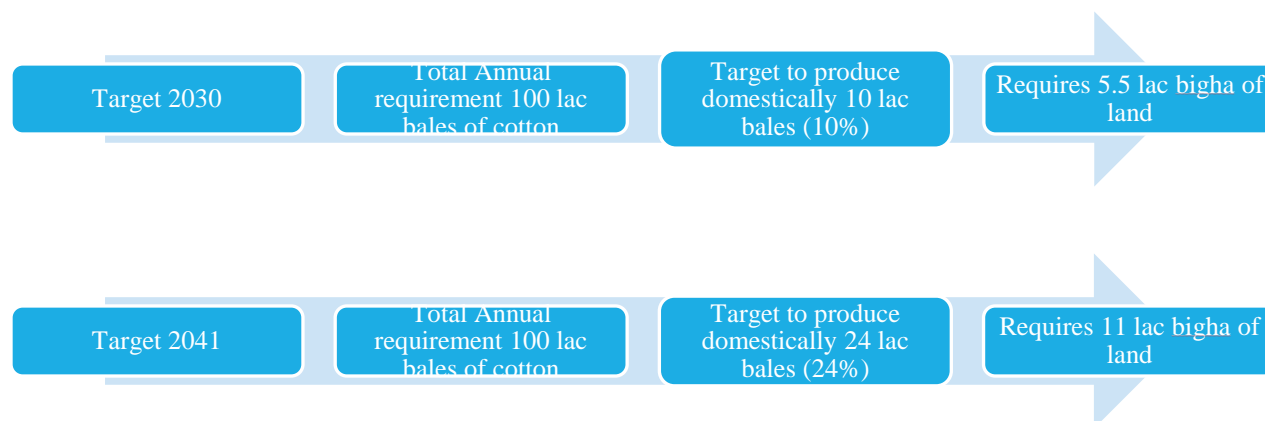


Fig-3: Targeted domestic raw cotton production of 10% by 2030 and 24% by 2041 to feed spinning mills in Bangladesh

This method of cultivation is called agroforestry. In Bangladesh there are about 5000 ha areas of land under agro-forestry; from that agroforestry land easily 2000 ha area could be taken under cotton cultivation [15]. Seed cotton production & conversion to raw cotton in bale is shown in figure-2. Targeted cotton production by year 2030 & 2041 is shown in figure-3.

### RESEARCH METHODOLOGY

This descriptive research was conducted to focus on the importance of cotton production and to identify different potential areas in Bangladesh for cotton production. To prepare this research study, the current practices of the garment and textile sectors were evaluated and practical experience and mixed method was used to identify the gap. So this paper is mainly prepared by thorough analysis of different documents and articles like export policy of 2021-2024, import policy of 2015-2018, rules and regulations of NBR (National Board of Revenue), books on organization development, lecture sheet, garment manufacturing industry annual report, BGMEA (Bangladesh Garment Manufacturers and Exporters Association) yearly report and files, different journals and newspapers etc. Finally, all the information from different sources was grouped together to produce this unique paper.

### RESULTS AND DISCUSSION

#### Cotton Production in Bangladesh

In order to achieve Bangladesh's goal of 50 billion apparel export within 2021 and further goal to take global leadership position in textile and apparel, sustainable and strong backward linkage Primary Textile (mainly spinning) Sector (PTS) is a must. As the textile industry is mostly cotton based industry, Bangladesh cannot think a single moment in the field of spinning industries without uninterrupted continuous supply of cotton fiber. As Bangladesh doesn't produce cotton much, the second largest cotton based RMG exporter country is almost totally depended on cotton import. In spite of being agriculture dependent country, Bangladesh can meet up only 1% of its cotton demand. Understanding the importance of cotton as cash crop, Bangladesh Cotton development board (CDP) is trying to improve the cotton cultivation scenario day by day.

In fiscal year 2016/17 though, planted area levels were unchanged still the production revised up by 5 percent to 125,000 bales. In 2017/18 also, cotton will be planted at 43,000 hectares (HA) unchanged, but production is forecast to rise to 130,000 bales as because of increasing use of the long staple American Upland variety. It can produce 1 million bales of cotton in 200,000 hectares of land adding that this can save 10 percent to 15 percent import costs. Bangladesh primarily produces American Upland (*Gossypium hirsutum*) and Tree (*Gossypium arboretum*) cotton that represent 95 and five percent of total production, respectively. Upland cotton is cultivated in the southwestern, northern, and central region, and tree cotton is grown in three southeastern hill districts. The average length of Upland cotton is greater than 28 millimeters (mm); Tree cotton is less than 10 mm.

Contacts believe American Upland cotton will be planted in hilly areas instead of other non-food crops. Bangladesh Cotton Development Board (CDB) received approval from the National Committee on Biosafety to initiate a contained trial of eight Bt Cotton hybrid varieties. Mahyco Seed Company Ltd. is supplying Bt cotton seed containing Bollgard II double Bt genes Cry1Ac and Cry2Ab. CDB is planning to start trials during the next cotton season (July 2017). Previously CDB

found that field trials conducted on Bt Cotton variety Bt hybrid HSC-4 produced unsatisfactory results [16].

### **Potential Cotton Production Region in Bangladesh**

#### ***Barendra Area:***

According to Bangladesh Cotton Development Board (BCDB), there are about 1, 50,000 hectares of unused land in Barendra area in North Bengal while 60,000 hectares are being used for tobacco cultivation. "Bangladesh can produce nearly 2.0 million bales of cotton locally which can meet one third of the demand the country has for the apparel industry. As the second largest consumer, Bangladesh consumes 61 lakh cotton bales, of which 1.5 lakh are produced locally. It would like to replace tobacco cultivation with that of cotton as the country has a huge demand for it. The cotton cultivation has no bad impact, which, on the other hand, exists in tobacco farming. The reason why farming cotton in northern area is advantageous is its geographical feature, i.e. char area, and dry weather, plus congenial atmosphere to agroforestry. We have a litany of farmers and the government is providing financial support for them." Long growing period, high input cost, climate change impact, especially erratic rainfall, price volatility, lengthy return of investment, lack of grading system and dependency on a handful of private cotton-ginners are challenges towards boosting cotton cultivation. Farmers present urged the government to invent such a variety of cotton that takes shorter period of time to grow and harvest, or else initiatives will end up in failure. The cotton development board is working on introducing a high-yield variety to bring benefits to the growers [17].

#### ***Rajshahi:***

Cotton production in the region is bright as its topography and climatic condition suitable for the cash crop, reports BSS. Cotton farming has been gaining popularity among farmers as they are getting more money from the cultivation than any other crop. Acreage of the crop is gradually increasing in the region comprising Rajshahi, Naogaon, Natore and partly in Pabna for the last couple of years. Farmers have started replacing paddy and sugarcane cultivation with cotton farming because it requires less investment and less labor. Most of the farmers cultivate varieties like CB-12 and three hybrid varieties- Rupli-1 and DM 1 and 2. Cotton Development Board help the cotton farmers in procuring seeds, making land, nurturing of crop. In last 2015-16 fiscal year, 6,736 bales were harvested from 145 hectares of arable land. Considering the demand of the country's textile industries, the local cotton growers can meet only 3-5% of the annual demand and the rest is met through import. [18].

#### ***Jessore:***

Cotton farmers can reduce import dependency. The prospect of cotton cultivation across the southwestern part of Jessore is giving new horizons. Cotton is grown in around the country from one and a half to two hundred thousand bales. The concerned people are expected to produce 10 lakh bales of cotton in the next 5 years. This will save the country's huge foreign currency in textile sector. Against this huge amount of imported cotton, the foreign currency (dollar) is counted in millions of dollars. Authorities claim, the yield of high yielding (hybrid) cotton cultivation is going to be more than doubling - which can be done by the farmers in the desired cotton house. These information was found from the responsible sources of the Bangladesh Cotton Development Board. It is known that about 13 thousand farmers have planted cotton in 21 units under the district of Jessore of Southwest District. Agricultural exhibitions continue to be free of agricultural fertilizers, fertilizers, irrigation, pesticides and other agricultural inputs.[19].

***Jhalakathi:***

Cotton cultivation is expanded in Jhalakathi. The field is filled with white cotton in the season. And it is going from the garden to the Chuadanga cotton mill. The price is too good to match. Cotton cultivation seems to be a new possibility for the farmers of this region. A cotton cultivation of Rupal 1 variety started in 2011, with the help of the cotton development board of Jessore, is now spreading in a little more than 5/6 years in the village of Dhanasiri Union of Sadarupazila. Many farmers have leaned cotton cultivation due to cotton prices being fairly good. A kind of Gulmatti tree is full of flowers in the Kartik month. From the flower to the month of Chaitra, the farmers collected cotton from the field. Big has produce 10 cotton cotton growers. New possibilities for farmers of cotton cultivation. There is less risk of losses. Jhalokathi has been helping farmers in cotton cultivation in various ways [20].

***Mymensingh:***

Cotton cultivation has increased in greater Mymensingh district. The target for cotton production has been set to 10 thousand 943 bales. The farmers of greater Mymensingh districts have become interested in cotton cultivation as a profitable crop. The training and materials provided by the Board for providing training to the turtles have been provided. These assistance to be helpful in increasing the yield. Due to favorable weather, cotton cultivation of more land has been done [21]

***Chittagong Hill Tracts:***

Cotton cultivation has become popular in Khagrachari and three hill districts. The offices of the Cotton Development Board have been established in Khagrachari. Cotton Development Board officials are encouraging farmers to cultivate cotton in scientific method. The cultivation of CB-12 and Rupal-type hybrid cotton are low and the yield is also good. Besides, it is sold at good prices. Cotton crops are also cultivated. The target is 150 to 200 metric tons. In addition to traditional varieties of zoom, they are cultivating hybrid varieties of cotton. Cotton production to be increased to 600 to 700 metric tons in the next five years. There is a demand for cotton in the country 42 lakh (five tons) in one sack. The possibility of loss of farmers in cotton cultivation is very low. [22].

***Kushtia:***

Farmers of Tacca have been dreaming to be financially benefited by hybrid cotton cultivation. The target of cotton cultivation in the greater Kushtia district has been estimated at 8, 190 hectares. It has been cultivated in 2,472 hectares of land, yielding high yielding hybrids has been cultivated on 385 hectares of land. Of these 815 hectares in Chuadanga district, 849 hectares in Meherpur district and 702 hectares in Kushtia district. Now the target for cotton production is 19 thousand tones. Kushtia zone region is particularly suitable for cotton cultivation. Due to lack of proper planning, the cultivators of cotton in the region have been left with other crops except cotton cultivation due to lack of proper yield and cotton prices. In the 1980s there was a lot of cotton cultivation in this region. Despite the vastly sown cotton cultivation, the cultivators dropped cotton cultivation. If the government determines the correct price than the international market, people in this region will be tilted in cotton. Modern agricultural scientists are inventing agricultural produce in the era of modern technology. But there is no better variety of cotton today, Not to touch any modernity. In the 1980s, no other species were invented except CB-9 varieties. If cultivate the same varieties repeatedly on the same soil, there is no good yield. 23].

### **Rangpur and Dinajpur:**

Cebi-14 and high yielding varieties of cotton cultivation are increasing in Dinajpur and Rangpur regions. Cotton cultivation is becoming increasingly popular as a companion crop. The possibility of cotton cultivation in the northern region of the country is inaccessible. Cotton cultivation along with basic crops in the land and weather in the country has increased. And if this cotton cultivation is increased, the import cost will be reduced every year. White Gold of Bangladesh could become an agricultural product. Currently cotton is being cultivated in the flat, mountainous and saline lands in the southern part of the country. However, newer crops are grown in the northern region of the country. It can be possible to produce Bari Mug-6, Lalmaras, Mardazak, Data, coriander leaves, almonds, sesame seeds, moongs, jute seeds, ghee, paddy, rice, banana and other crops as a cotton crop. It is possible to produce two crops in the same crop and earn more profit. Recently, the yield of cotton has increased manifold due to the introduction of hybrid and high yielding varieties of cotton. With that, cotton cultivation has now become a profitable crop because cotton market has increased. Presently, the second largest cash crop of cotton and the main raw material of textile industry. Plant cultivation of plain land is being done in 34 districts of the plain area and most recently, there are hill species in the three hill districts as well as cultivation of plain land cotton cultivation. The Cotton Development Board is continuously working with cotton farmers and is going to expose the possibility of day by day. Cotton is one of the most profitable crops in Bangladesh today. village of Biralupazila of Dinajpur. He said, he has cultivated cotton in 2 bighas of land. As the country's climate and soil are suitable for cotton cultivation, initiatives have been taken to promote cotton cultivation throughout the country. Our country is engaged in cotton cultivation[24]

### **Climate Change and Mitigation Strategy**

Cotton covers about 2.5% of the world's arable lands, and related to 0.1% to 0.3% of global GHG emissions. It is therefore not a principal source of GHG emissions. Yet cotton can contribute to mitigating climate change, in particular by increasing efficiency and reducing emissions from the more efficient use of carbon based fuels and inputs made therewith (irrigation water, fertilizers, pesticides, etc and adoption of low input and organic practices. On a field level, the following mitigation measures can be identified in order to increase cotton crop efficiency in terms of yield per unit of GHG emitted:

- Minimize soil tillage on cotton cropland in order to prevent soil to air emissions;
- Minimize carbon-based fuel mechanization and transport;
- Minimize the use of synthetic fertilizers in general and nitrogen fertilizers in particular, because these are an important source of N<sub>2</sub>O emissions;
- Minimize the use of irrigation water, because of its carbon-based fuel footprint, and reduce competition for freshwater for man and nature;
- Minimize the use of industrial preparations such as pesticides, herbicides and defoliant because of their carbon fuel footprint;
- Minimize the burning of cotton crop residues where still applied, and recycle these for soil fertility management when not used as a fuel for cooking and heating;
- Adopt where feasible organic farming practices [11]

### **Cotton in the Income Generation of Tribal People**

Hill Cotton/Comilla Cotton is a short staple length, course fiber grown in three hill districts as Jhum crop, which has very good export value. Jhum is a mixed crop cultivated mainly in hill slopes where more than two crops are seeded in a pit and harvested sequentially. In general, 4-5 years interval is needed for further cultivation in the same land. The main crops cultivated in Jhum are Aus rice,

Sesame, cotton, maize, marpha, chili and pumpkin. In three hill districts, huge area of land is under Jhum cultivation. If we can improve the variety and efficient management practice of cotton grown in hill area, economic development and improved livelihood of tribal farmers is possible. Cotton is only cash crop for tribal farmers. In this area, we need to address on a urgent basis so tribal farmers can go for better income generation [25].

### **Role of Cotton in Employment Generation Employment Generation in Spinning and RMG Sector**

Our textile sector through spinning and RMG has been creating employment for 5 million people of which 80% are women. So cotton as well as Textile sector can also contribute in generating employment. In addition, through marketing and ginning of cotton can also create employment opportunity in the rural and urban area.

Employment generation at farmer's level through cotton production: Presently, cotton is growing in 33 districts of Bangladesh and 35 thousand farmers are involved in production process from sowing to yield which is developed as the best employment opportunity in Bangladesh. As cotton is now a profitable crop so more farmers are increasingly interested to grow cotton. If we can increase cotton production and area for cotton production, the number of farmers will be increased and more people will get involved eventually [25].

### **Role of Cotton in Employment Generation Employment Generation in Spinning and RMG Sector**

Our textile sector through spinning and RMG has been creating employment for 5 million people of which 80% are women. So cotton as well as Textile sector can also contribute in generating employment. In addition, through marketing and ginning of cotton can also create employment opportunity in the rural and urban area. Employment generation at farmer's level through cotton production: Presently, cotton is growing in 33 districts of Bangladesh and 35 thousand farmers are involved in production process from sowing to yield which is developed as the best employment opportunity in Bangladesh. As cotton is now a profitable crop so more farmers are increasingly interested to grow cotton. If we can increase cotton production and area for cotton production, the number of farmers will be increased and more people will get involved eventually.

### **Employment in Private Ginning Industry**

Ginning is the process of separation of fiber and seed from seed cotton. Farmers harvest seed cotton from the cotton field. The private ginning industry is involved to purchase seed cotton from the farmers through local traders and separate seed and fiber through ginning process and send the fiber to spinning mill by making bale and seed is used for different value addition product like edible oil, cotton oil cake for animal and fish feed, used in soap industry etc. For the processing of seed cotton many workers mostly women are employed. Expeller also is used for oil extraction from seed as crude oil after refine is being used as edible oil. Many men and women are used as workers in these industries. So if the cotton production is increased the number of ginning industry will be increased and scope of employment generation will be increased [25]

### **Profitability of Cotton**

Cotton is a highly competitive and long duration crop, it requires 6 months from sowing seeds to yield cotton. Within this time our farmers can grow two crops from the same land through which he can generate more income from same land. High and medium high land is suitable for cotton production where many high value crops can grow. During early and mid-ninety's, the area and

production was increasing but from late ninety's the acreage and production was drastically dropped. During that time cotton price was also low in international market. So, cotton was not profitable for most of the farmers. That's why the farmers were migrating to grow other short duration high value crops like vegetables, wheat, maize, potato, banana, flower etc.

## CONCLUSION

Bangladesh have to increase cotton production to keep sustainable Ready-Made Garments sector but of course it has to be accomplished by keeping food security intact. Cultivation can be further boosted if government encourages cotton cultivation despite tobacco cultivation. As a cash crop cotton can be good alternative to tobacco production for the farmers. According to Md. Abu Taleb Chowdury, Fiber Technologist, Cotton Development Board, "cotton development board is more capable than before to face challenges of cotton cultivation. Our scientists are trying to invent water tolerable cotton species; they are also working to develop cotton species convenient for hill track area to boost up our cotton cultivation. Actually, land scarcity is main problem for us. Our government's priority is to maintain food securities as well as we need to increase cotton cultivation. But there is no proper coordination between Agricultural ministry & Textile ministry. Proper coordination and adequate training facilities among cotton farmers can change the whole scenario of cotton cultivation."

Cotton provides food, feed, fiber and fuel. It contributes in food security by increasing by purchasable power of the farmers. It creates income and employment, which has significant impact on economic and social aspect. Cotton plays a key role in the national economy of different countries that is why it is called White Gold for many countries of the world. Government and other policy makers should develop positive support to boost up the cotton production.

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## Right Adrenal Myelipoma: Case Report

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### **Abstract:**

Adrenal myelolipoma is a rare benign tumor originating from the adrenal cortex. Composed of adipose tissue and bone marrow. The incidence of adrenal myelolipoma is 0.08 to 0.2% of autopsy cases and 0.06% of patients operated. Large tumors of a size greater than 7 cm should be removed. We report here the case of a 41 years old patient with a large right adrenal myelolipoma. The radiological outcomes and surgical management.

*Keywords: adrenal; myelolipoma; benign; mass; abdominal MRI*

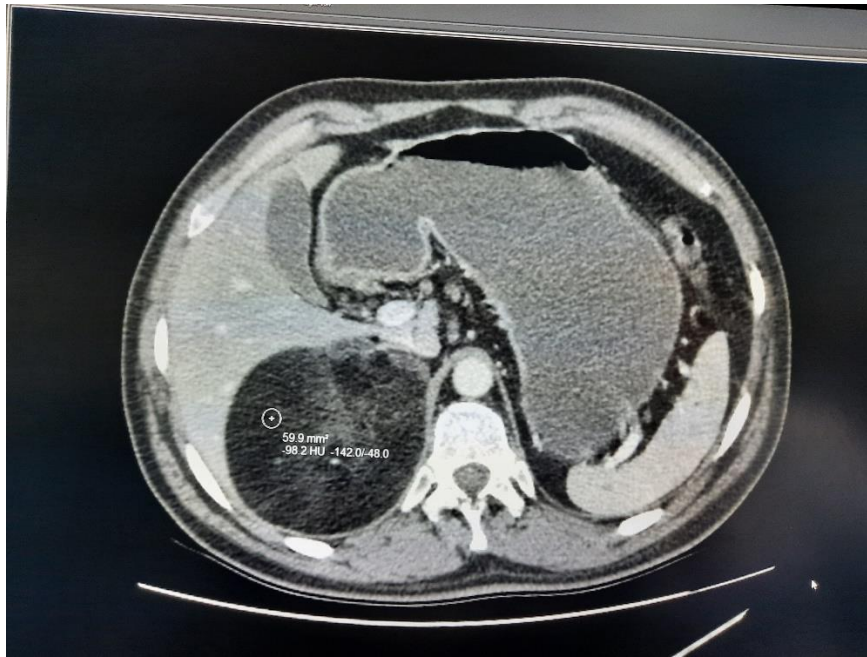
### **INTRODUCTION**

Adrenal myelolipoma is a rare benign tumor composed of variable proportions of adipose tissue and bone marrow represented by all three lineages of hematopoietic elements [1]. The pathogenesis of myelolipoma is obscure, usually asymptomatic and generally has a diameter <5 cm [2]. Its imaging characteristics are so specific that the radiological diagnosis can be certain [3]. Large tumors require surgical treatment [3].

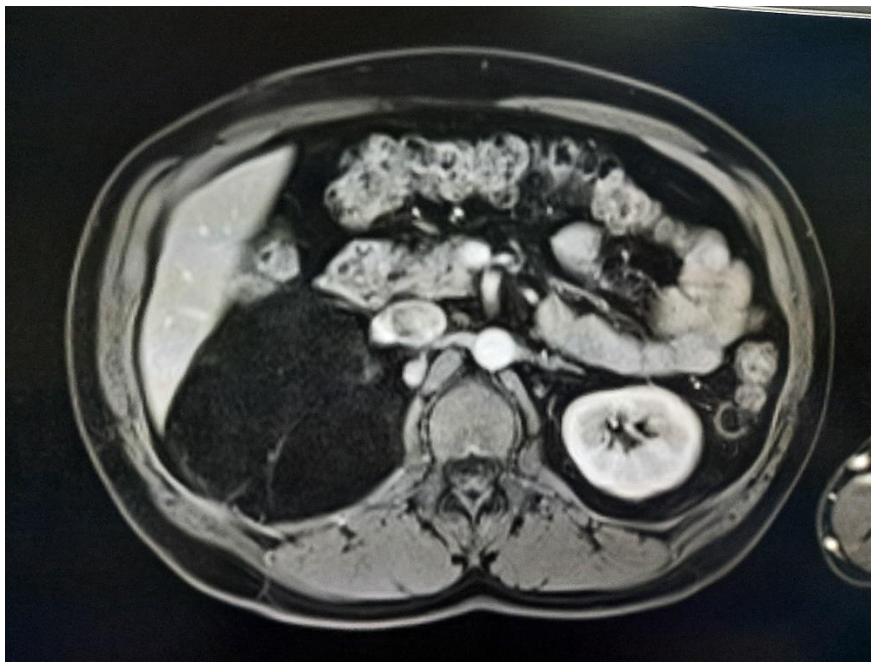
### **CASE PRESENTATION**

A 41 years old male patient, chronic active smoker, presented with chronic right lower back pain for two years, without hematuria, neither lower urinary tract disorders or associated digestive symptoms. The whole evolving in a context of apyrexia and conservation of the general status. On clinical examination, the patient was conscious 15/15, with normal blood pressure 120/70 mmHg, normal heart rate 72 bpm and apyretic. The urogenital examination showed tenderness in the right lumbar fossa.

On the paraclinical level, an uro-TDM was performed showing a rounded mass on the adrenal space, with regular contours, fat density (-90 HU), with thin septa, measuring 98x95x94 mm (figure: 1). An abdominal MRI was performed in addition, showing an oval right adrenal mass, in T1 and T2 hypersignal, cancelling in FAT SAT without any sign of loco-regional invasion (figure: 2). This radiographic appearance was in favor of an adrenal myelolipoma. The biological examination was without abnormalities.



**Figure 1: Uro-TDM showing the mass**



**Figure 2: MRI with oval right adrenal mass**

Given the tumor size and the painful symptomatology of the tumor mass, surgical exploration by thoraco-phreno-laparotomy was performed. The exploration showed a large right retroperitoneal mass without signs of invasion of adjacent organs. A tumorectomy was performed without incident. The postoperative course was simple. The histopathological examination was in favor of an adrenal myelolipoma.

### **DISCUSSION**

Adrenal myelolipoma is a rare benign tumor originating from the adrenal cortex and is usually nonfunctional[1]. This neoplasm is composed of variable proportions of adipose tissue and bone marrow represented by all three lineages of hematopoietic elements.

The incidence of adrenal myelolipoma is 0.08 to 0.2% of autopsy cases and 0.06% of patients operated. The average age of onset is 54 years with a slight male predominance. It is most often unilateral and right-sided, however, bilateral forms have been described[2].

The pathogenesis of myelolipoma is obscure. It is hypothesized that the primary event is a metaplastic change occurring in the reticuloendothelial cells of the blood capillaries due to stimuli like necrosis, infection or stress. Another major group of hypotheses relates to hormonal pathways (adrenocorticotropin hormone)[3].

Adrenal myelolipoma is usually asymptomatic and generally has a diameter <5 cm, discovered either during an autopsy or, more often, during radiological investigations for other conditions. A high blood pressure may be observed in the case of compression of a renal artery[2].

A small percentage of patients presents with abdominal symptoms, flank pain or haematuria. Even life-threatening haemorrhagic shock has been described in myelolipomas larger than 10 cm. The symptoms appear to be caused by mechanical compression or tumour necrosis or retroperitoneal haemorrhage[4].

Its imaging characteristics are so specific that the radiological diagnosis can be certain. The echography shows a heterogeneous lesion with a hyperechoic aspect corresponding to the fatty contingent. The computed tomography shows a mass with regular contours, with two components : the mature adipose tissue spontaneously hypodense (< 30 HU), and hematopoietic marrow cells (30-50 HU), heterogeneously enhancing[5]. On magnetic resonance imaging, the appearance of the tumor demonstrates the high signal intensity and reduced signal intensity depending on the T1-weighted or T2-weighted sequences, respectively. Retroperitoneal fat-containing tumors like teratoma, lipoma, myolipoma, angiomyolipoma, and liposarcoma may mimic AML radiologically[1].

Small tumours should be managed by annual radiological screening in an outpatient setting. Large tumors of a size greater than 7 cm should be removed, as they carry a potential risk of malignancy and retroperitoneal haemorrhage due to spontaneous rupture[6].

Symptomatic tumors or with hormonal activity should also be removed by laparotomy or laparoscopy. Laparoscopic adrenalectomy is safe and effective [3] Larger tumors require greater exposure, and it may be necessary to make large incisions such as Chevron (bilateral subcostal) or thoracoabdominal incisions as our case. Good exposure is crucial to avoid damaging the vena cava on the right or the aorta on the left, among other structures. The fact that myelolipomas are encapsulated allows them to be easily separated from the surrounding structures[7].

## CONCLUSION

Adrenal myelolipoma is a benign and nonsecretory tumor that is often discovered incidentally. Its size can vary from a few millimeters to tens of centimeters; it can compress neighboring organs, becoming symptomatic. The preoperative diagnosis can be established by medical imaging to avoid unnecessary surgery in small asymptomatic tumors. In other cases, a surgical removal is necessary to demonstrate benignity.

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# Determinants of Diarrhea Among Under-Five Children-Visiting Government Health Facilities in Nekemte Town, Western Ethiopia: Unmatched Case Control Study

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## Abstract:

**Background:** Diarrheal disease is the most common cause of illness and the second leading cause of child death next to pneumonia in the World. The aim of the study was to identify the determinants of diarrheal diseases among under five children visiting government health facilities in Nekemte Town, Western Ethiopia. **Method:** Facility based unmatched case-control study was conducted in Nekemte Town Government Health facilities. Four hundred seventy-seven (159 cases and 318 controls) under five children were selected using consecutive sampling method. Data were cleaned, coded and entered in to Epi data version 3.1 then exported to SPSS version 20 for statistical analysis. Association between dependent and independent variables were computed using multivariable logistic regression and significance of the associations were declared by p-values with their corresponding 95% confidence interval along with adjusted odds ratios. **Result:** A total of 447 (159 cases and 318 controls) were participated with equal response rate of 100%. In this study: diarrheal morbidity is significantly determined by family income [AOR: 0.435, 95% CI: (0.263-0.72)], supplementary feeding commenced time [AOR: 5.38, 95% CI: (3.23-8.94)] and Measles vaccination [AOR: 3.72, 95% CI: (1.6.9-8.63)] respectively. **Conclusion and Recommendation:** In this study family income, Supplementary feeding commenced time and measles vaccination status were determinants of diarrhea morbidity among under-five of age visiting public health facilities in Nekemte town. Health service providers should aware mother/caregiver on supplementary feeding commenced time and measles vaccination.

**Keywords:** Unmatched Case-control, Under Five Childhood Diarrhea Morbidity, Ethiopia

## INTRODUCTION

Diarrhea is a major health problem and usually a symptom of an infection in the intestinal tract, which has a variety of causative agents including viruses, bacteria and parasites. The infection spreads through the ingestion of contaminated food or drinking-water, or person-to-person as a result of poor hygiene. There are three clinical types of diarrhea: Acute watery diarrhea which lasts several hours or days and includes cholera; acute bloody diarrhea (dysentery); and persistent diarrhea that lasts 14 days or longer [1-3].

Diarrhea is a leading cause of morbidity and mortality across all age groups and regions of the world. Though mortality rates among 9-12 years of children, adolescents, and adults are lower than those observed in children under five, diarrhea still poses a substantial burden accounting

for approximately 2.8 billion diarrhea episodes among older children, adolescents, and adults [4-6].

Diarrheal disease due to unsafe water and lack of sanitation is the greatest cause of morbidity and mortality in under-five children in the world, especially in poor countries. Diarrhea alone kills more children than AIDS, malaria, and measles combined [4-6].

It is estimated that 6,000 children die every day from diarrheal diseases alone and large proportion of diarrheal disease in the developing world are due to poor water-handling practices, sanitation and hygiene. A child dies every 15 seconds from diarrhea caused largely by poor sanitation and a contaminated water supply [7, 8].

As a child's immune system is progressively compromised with each attack of diarrhea, related illnesses kill millions or more indirectly. Almost 90% of diarrhea is attributed to unsafe drinking water, inadequate sanitation and poor hygiene [7, 8].

Many children in the developing world cannot access urgent medical care for severe illnesses, making prevention methods, including improved hygiene, sanitation, safe drinking water, exclusive breastfeeding, and also rotavirus vaccines which are a critical component of diarrheal disease prevention and control. When diarrhea occurs, it can be successfully treated with simple solutions, including oral rehydration therapy/oral rehydration solution (ORS), zinc and other micronutrients, and continued feeding [9, 10].

Diarrheal disease is the second leading cause of death in children under five years old, and is responsible for killing around 760,000 children every year. Diarrhea can last several days, and can leave the body without the water and salts that are necessary for survival. Globally diarrhea accounts for 17% of the deaths and 5% of health loss to disability among children under the age of five [11, 12].

Most people who die from diarrhea actually die from severe dehydration and fluid loss. Worldwide; there are 2.5 billion cases of diarrhea each year among under five children which is likely to result in death or other severe outcomes. More than half of these cases occur in Africa and South Asia. About 80% of deaths are still in Africa including Ethiopia and the average annual incidence rate of diarrhea in under-fives is estimated to be 2.6 episodes [11, 12].

Diarrheal diseases account for 1 in 9 child deaths worldwide. It kills 2,195 children every day more than AIDS, malaria, measles and 801 thousand Child deaths every year losing nearly 32 school buses full of children each day. Despite these sobering statistics, strides made over the last 20 years have shown that, in addition to rotavirus vaccination and breastfeeding, diarrhea prevention focused on safe water and improved hygiene and sanitation is not only possible, but cost effective: every \$1 invested yields an average return of \$25.50. A child dies every 15 seconds from diarrhea caused largely by poor sanitation and contaminated water supply [13, 14].

Water sources and sanitation facilities have an important influence on the health of household members, especially children. Water safety in a community at large and household levels depends on a range of factors to prevent childhood diarrhea, the quality of source water to storage, throwing away disposal of feces, point-of use household water treatment, washing of containers before transferring, duration of stored water, cover of container during transportation and

storage, unhygienic water drinking and handling in the domestic setting [15].

The consequences of diarrheal diseases on childhood are huge; leading to decreased food intake and nutrient absorption, malnutrition, reduced resistance to infection, and impaired physical growth and cognitive development [16, 17]. The cause of under five deaths in Ethiopia due to diarrhea are 23%. Under five mortality rates are 88 and 112 per 1000 lives birth in Ethiopia and Oromia respectively. [18, 19]

In Ethiopia, morbidity reports and community-based studies indicate that diarrheal diseases are public health problem that causes excess morbidity and mortality among under five children. However, various community-based studies conducted on different parts of Ethiopia indicated that the prevalence of diarrhea among under five years age vary for instance in Arba Minch district 30.5%, Nekemte town 28.9%, Mecha district 18% [20, 21].

According to Nekemte Town Health Office report diarrheal disease is one of the major public health problems in the area and it is among one of the top ten diseases causing morbidity and mortality in under-five children. There are few institution-based studies conducted on different parts of Ethiopia and significant number of patients coming to government Health Facilities in Nekemte town with diarrhea complaint. Therefore, the objective of this study was to identify potential determinants of acute childhood diarrhea among under five children.

## METHODS

### **Study Design, Setting, and Population**

The study was conducted in Nekemte town, East Wollega Zone, Oromia regional state, Western Ethiopia. Facility based Unmatched case control study was used.

#### **Source Population:**

The source populations were all under-five children visiting government health facilities of Nekemte town.

#### **Study Population:**

The study populations were selected children less than five years of age visiting government health facilities in Nekemte town who were included in the actual data collection.

#### **Cases:**

Under five children with diarrhea visiting government Health Facilities in NekemteTown come for treatment at outpatient department.

#### **Controls:**

Under-five children without diarrhea visiting government Health Facilities in Nekemte Town come for treatment of other cases at outpatient department.

#### **Eligibility Criteria:**

**1. Inclusion Criteria:** Children of under five age with mothers/ caregiver attending government health facilities in Nekemte town at under five outpatient department during the study period.

**Cases:** were under five children with diarrhea visiting government Health Facilities in Nekemte Town come for treatment at outpatient department.

**Controls:** were under-five children without diarrhea visiting government Health Facilities in

Nekemte Town come for treatment of other cases at outpatient department.

2. **Exclusion Criteria:** Mothers/caregivers of under five children unable to hear or speak, and those mothers or caregivers with under- five urgent referral cases, at outpatient department for both cases and controls.

## Sample Size Determination and Sampling Techniques

### **Sample Size Determination:**

The required sample size is calculated by double population proportion formula for unmatched case control study using Epi Info version 7 Stat Calc function. From similar study conducted in Kotebe sub city, Addis Ababa, hand washing practice taken as the main predictor of outcome (diarrhea).  $P_1$  = proportion of cases exposed to hand washing after cleaning child's buttock was 0.427,  $P_2$  = proportion of control exposed to hand washing after cleaning child's buttock was 0.571[25].

**Table 1. Associated variables for sample size determination.**

Associated Variables	% of control exposed or $p_2$	% of Cases Exposed or $P_1$	Confidence interval	Power	Odds ratio	Sample size + 5% non-response rate				References
						Cases	Controls	controls to cases ratio	Total	
Maternal education	47.6	23.1	95%	80%	0.33	51	102	2:1	161	Aklilu T. & Zewdie A., 2014
Supplementary feeding commenced	43.2	29.1	95%	80%	0.3	148	296	2:1	452	
Hand Washing after cleaning child's buttock	57.1	42.7	95%	80%	0.59	159	318	2:1	477	

Then the largest sample size was selected for this study as follows

Case = **152**

Control = **303**

Total = **455**

Non response rate = **5%**

Total Cases = **159**

Total Controls = **318**

Final Sample size = **477**

### **Sampling Techniques:**

One hospital and two health facilities are included in this study. Then the calculated sample size was distributed to the health facilities using proportional to size based on total under five visited at OPD in previous year to assure representativeness. In this allocation, total number of under five visited at OPD in each government Health facilities during the previous year from July 8, 2015 to July 7, 2016 which are 14068 Nekemte Hospital, 2173 Nekemte health center and 645 Chalelike Health center were used. Accordingly, 83% (396), 13% (63) and 4% (18) of the sample were allocated for Nekemte Hospital, Nekemte Health center and Chalilake Health center respectively. The survey was conducted in under five oPD to identify, register cases and controls. Cases and controls were selected by consecutive sampling from each facility based on fulfillment of case definition criteria.



## Study Variables

### **Dependent Variables:**

Childhood Diarrhea morbidity status.

### **Independent Variables:**

1. **Socioeconomic & Demographic Status:** family income, place of residence, maternal age, education, ethnicity, number of children, occupation, marital status, religion.
2. **Behavioral Factors:** hand-washing, method of water drawing and storage, infant and young child feeding practices, duration of breast-feeding, time of introducing supplementary feeding, child immunization.
3. **Environmental Factors:** type of water source, distance to the water source, amount of daily water consumption availability of latrine, number of rooms, livestock in house, and refuse disposal.

## Operational Definitions

**Diarrhea:** is defined as having three or more loose or watery stools per twenty-four hours in two weeks period preceding the data collection, as reported by the mother/care taker of the child.

**Index Child:** refers to a child that was included in the study from a household to have information on the demographic and health characteristics.

**Improved Water Source:** Water from protected springs and/or wells, from pipe and from distribution post unless considered as unimproved

**Number of Rooms:** are defined as any partition of a house that is intended to separate the rooms for different purposes.

**Refuse:** includes such solid wastes as ash, cow dung, home-sweepings; but not human excreta.

**Exclusively Breast-Fed:** A child who was receiving no food, solid or liquid, other than breast milk at the time of the survey.

**Mixed Feeding:** a child who was receiving food, solid or liquid, in addition to breast milk at the time of the survey.

## Data Collection Procedure

### **Data Collection Instrument:**

Data collection tool were adapted from various similar studies in different parts of the world and modified according to local context. Data collection tool is translated from English to local language, "Afan Oromo" then, back translated to English to check for consistency by different person.

### **Data Collection Techniques:**

Data were collected by interview using interviewer administered structured questionnaire. Data collectors were college nursing students and supervisors were nurses who work in that health facility. There were 6 data collectors, 2 supervisors and the overall activities were supervised by principal investigator.

**Data Quality Control:**

To assure the quality of data, the following measures were undertaken pre-testing of the questionnaire, the final version of the questionnaire was translated into the local language of the respondents (Afan Oromo), and data collector and supervisors were trained for two days by the principal investigator on the objective of the study, the methods of data collection, how to recruit cases and controls, and data collectors were familiarized with data collection tools with respect to the study with practical exercises.

Pretest was conducted in neighboring Sibu Sire health centers which is 50km from the study area. By taking 5% of the sample size that were not included in the actual study population for three days before the actual data collection takes place. The collected data were checked for its completeness at the end of interview and at the end of the day by principal investigator. Missing values and outlier were checked before analysis by running descriptive analysis.

**Data Processing and Analysis**

Data completeness were checked manually, then entered into Epi-data software and exported to SPSS Version 20 for analysis. Descriptive statistics was used to describe frequency and percentage of diarrhea morbidity among under five children. Bivariate analyses were performed to nominate candidate variables for multivariable analysis with p-value less than 0.25. Multivariable logistic regression was used to identify independent factors associated with diarrhea morbidity. The assumption fitness was test by Hosmer Lemeshow goodness fit test. Independent factors associated with diarrhea morbidity were declared with P-value less than 0.05 at 95% CI as cut of point.

**RESULTS****Socio-Demographic and Economic Characteristics**

A total of 159 cases and 318 controls were included in the study and in both group the response rate was 100%. Among cases 82 (51.6%) and 177 (55.7%) controls were males respectively. Concerning the age groups 55 (34.6%) of cases and 130 (40.9%) controls were in the age groups of 24-59 months. Of total cases 85 (53.5%) and 170 (53.5%) controls were had 3-5 family size. From total cases 72 (45.3%) and controls 161 (50.6%) were protestant. Regarding ethnicity 142 (89.3%) and controls 286 (89.9%) were Oromo.

Out of total 41 (25.5%) cases and controls 83 (26.1%) mothers/caregivers and 71 (44.7%) cases and controls 139 (43.7%) fathers had Higher Education. From total cases 105 (66.1%) and 201 (63.2%) controls mothers/caregivers and 76 (47.8%) cases and control 144 (48.3%) fathers were housewives and government employers respectively. Of total cases 148 (93.1%) and controls 291 (91.5%) were married. Regarding monthly income 81 (50.9%) cases and 195 (61.3%) controls earn greater than 1000 ETB.

In Bivariate analysis from all socio demographic factors only Age of child and family income were nominated with p-value  $\leq 0.25$  for multivariable logistic regression of under-five age childhood diarrhea morbidity.

**Table 2. Bivariate analysis socio-demographic and economic characteristics of mothers/care givers children under 5 years of age in Nekemte town**

Variables	Category	Cases (n=159) No. (%)	Controls (n=318) No. (%)	COR (95% CI)	P-values
Age of child (months)	<6	16 (10.1)	40 (12.6)	1.05 (0.54-2)	0.868
	6-11	33 (20.8)	73 (23.0)	0.93 (0.5-1.57)	0.802
	12-23	55 (34.6)	75 (23.6)	0.57 (0.3-0.92)	0.22
	24-59	55 (34.6)	130 (40.9)	1	
Sex of child	Male	82 (51.6)	177 (55.7)	1	
	Female	77 (48.4)	141 (44.3)	2.15 (0.57-1.2)	0.398
Family size	1-2	6 (3.8)	12 (3.8)	1.00 (0.36-2.7)	1.000
	3-5	85 (53.5)	170 (53.5)	1.00 (0.67-1.4)	1.000
	>5	68 (42.8)	136 (42.8)	1	
Religion	Orthodox	67 (42.1)	117 (36.8)	0.89 (0.4-1.63)	0.717
	Muslim	20 (12.6)	40 (12.6)	0.78 (0.5-1.17)	0.236
	Protestant	72 (45.3)	161 (50.6)	1	
	Oromo	142 (89.3)	286 (89.9)	1	
Ethnicity	Amhara	13 (8.2)	25 (7.9)	0.95 (0.47-1.92)	0.897
	Others	4 (2.5)	7 (2.2)	0.86 (0.25-3.0)	0.825
	Illiterate	40 (25.2)	70 (22.0)	0.86 (0.5-1.48)	0.597
Maternal education	Able to read and write	21 (13.2)	50 (15.7)	1.76 (0.65-2.2)	0.615
	Primary (1-8)	27 (17.0)	52 (16.4)	0.95 (0.52-1.7)	0.87
	Secondary (9-12)	30 (18.9)	63 (19.8)	1.03 (0.58-1.8)	0.9
	Higher Education	41 (25.5)	83 (26.1)	1	
Occupation of Mother	House wife	105 (66.1)	201 (63.2)	1	
	Government Employee	42 (26.4)	70 (22.0)	0.87 (0.55-1.3)	0.546
	Merchant	8 (5.0)	30 (9.4)	1.95 (0.86-4.4)	0.106
	Daily labourer	4 (2.5)	17 (5.3)	2.22 (0.72-6.7)	0.161
Occupation of Father	Government Employee	76 (47.8)	144 (48.3)	1	
	Merchant	28 (17.6)	60 (18.9)	1.18 (0.6-2.03)	0.535
	Farmers	36 (22.6)	73 (23.0)	1.04 (0.6-1.69)	0.869
	Daily labourer	19 (11.9)	41 (12.9)	0.91 (0.51-1.6)	0.768
	Illiterate	22 (13.8)	40 (12.6)	0.92 (0.5-1.68)	0.807
Parental education	Able to read and write	27 (17.0)	52 (16.4)	0.98 (0.57-1.6)	0.953
	Primary (1-8)	15 (9.4)	39 (12.3)	0.98 (0.57-1.6)	0.400
	Secondary (9-12)	24 (15.7)	48 (15.1)	1.02 (0.57-1.8)	0.941
	Higher Education	71 (44.7)	139 (43.7)	1	
Marital status	Married	148 (93.1)	291 (91.5)	1	
	Single	2 (0.6)	7 (2.2)	0.71 (0.2-2.28)	0.568
	Others	6 (3.8%)	20 (6.3%)	1.69 (0.6-4.31)	0.268
Monthly income	Less than 500	44 (27.7%)	51 (16%)	1	0.001
	501-1000	34 (21.4%)	72 (22.6%)	0.46 (0.29-0.7)	0.954
	>1000	81 (50.9%)	195 (61.3%)	1.01 (0.6-1.70)	

### Environmental Exposure Characteristics

From total cases 387 (81.1%) and controls 263 (68%) in the area were used pipe as main source of water. of total cases 121 (79.6%) and controls 241 (76.8%) had privately owned latrine. Out of total 152 (95.6%) cases and 314 (98.7) controls had hand washing availability within the Latrine. Of total cases 97 (61%) and controls 163 (51.3%) daily water consumption were  $\leq 20$  Liters/day. Concerning household solid wastes disposal method from total 74 (46.1%) cases and 165 (69.6%) controls used burning as main disposal of refuse

In Bivariate analysis from all environmental factors only daily water consumption and hand washing availability within the Latrine nominated with p-value  $\leq 0.25$  for multivariable logistic regression of under-five age childhood diarrhea morbidity.

**Table 3. Bivariate analysis of environmental characteristics of mothers/care givers children under 5 years of age in Nekemte town**

Variables	Category	Cases (n=159) No. (%)	Controls (n=318) No. (%)	COR (95% CI)	P-values	
Water source	Spring	35 (22%)	55 (17.3%)	0.74 (0.4-1.2)	0.215	
	Piped water	124 (78%)	263 (68%)	1		
Latrine Owned	No	38 (20.4%)	73 (23.2%)	1.18 (0.7-1.8)	0.48	
	Yes	121 (79.6%)	241 (76.8%)	1		
Latrine Availability	Yes	152 (95.6%)	314 (98.7%)	1		
	No	7 (4.4%)	4 (1.3%)	0.2 (0.08-0.9)	0.043	
Daily water consumption	<=20 Liters	97 (61%)	62 (39%)	0.6 (0.45-0.9)	0.044	
	>20 Liters	163 (51.3%)	155 (48.7%)	1		
Hand Washing Availability within the Latrine	No	7 (4.4%)	4 (1.3%)	0.2 (0.08-0.9)	0.043	
	Yes	152 (95.6%)	314 (98.7)	1		
Household solid wastes disposal method	Open field	Yes	73 (45.9%)	146 (45.9)	1.0 (0.68-1.4)	1.00
		No	86 (54.1%)	172 (54.1%)	1	
	Pit	Yes	42 (26%)	97 (30.5%)	1	
		No	117 (73.6%)	221 (69.5%)	0.8 (0.53-1.2)	0.355
	Burning	Yes	74 (46.1%)	165 (69.6%)	1	
		No	87 (53.9%)	153 (48.1%)	0.7 (0.5-2.03)	0.174
Garbage can	Yes	43 (27%)	70 (22%)	1.3 (0.84-2.0)		
	No	116 (73%)	248 (78%)	1		

### Behavioral Characteristics

From total cases 78 (49.1%) and controls 78 (49.1%) were those mothers/caretakers who started supplementary feeding for their children before the child reached 6 months and on 6 months respectively. Of total case 105 (72.4%) and controls 214 (77.5%) mothers/cares breast feed their greater than two years.

Out of total cases 136 (85.5%) and controls 292 (91.8%) had Separate can taking drinking water from storage. Of total cases 141 (90%) and 259 (81.5%) had complete vaccination. Only 58 (36.5%) cases and controls 93 (29.2%) used Pouring as method of drawing of water from storage container. And also, only 58 (38.4%) cases and controls 101 (29.2%) used dipping as method of drawing of water from storage container.

Of total only 14 (8.8%) cases and 13 (4.1%) controls had mother / caretaker diarrhea history. All most all cases 154 (96.9%) and 296 (93.1%) controls mother / caretaker were cut their child's finger nail well as their finger nail when it grows. From total cases 77 (48.8%) and controls 174 (69.3%) mother / caretaker were used both water and soap for hand washing.

From total cases 131 (82.4%) and 256 (80.5%) controls mother / caretaker were used cup and spoon for feeding their child. Only 40 (25.2%) cases and 55 (17.3%) controls mother / caretaker were used bottle for feeding their child. Of total cases majority of cases 97 (61%) mother / caretaker wash their hand after visiting latrine, whereas the majority of controls mother / caretaker wash their hand before food preparation and eating.

In bivariate analysis from all behavioral characteristics factors Supplementary feeding commenced time, Presence of Separate can take drinking water from storage, Measles vaccination, Pouring, Mother/caretaker diarrhea history and bottle feeding were nominated with nominated with p-value  $\leq 0.25$  for multivariable logistic regression of under-five age childhood diarrhea morbidity.

**Table 4. Bivariate analysis behavioral characteristics of mothers/care givers children under 5 years of age in Nekemte town**

Variables	Category	Cases (n=159) No. (%)	Controls (n=318) No. (%)	COR (95% CI)	P-values		
Supplementary feeding commenced time	On 6 months	62 (39%)	189 (59.4%)	1	0.000		
	Before 6 months	78 (49.1%)	43 (13.5%)	5.5 (3.4-8.84)			
	After 6 months	15 (9.4%)	53 (16.7%)	6.4 (3.2-12.6)			
	Not started	4 (2.5%)	33 (10.4%)	14.9 (4.9-45)			
Breast feeding status	<24 month	40 (27.6%)	62 (22.5%)	0.76 (0.4-1.2)	0.244		
	>=24 month	105 (72.4%)	214 (77.5%)	1			
Presence of Separate can take drinking water from storage	Yes	136 (85.5%)	292 (91.8%)	1	0.035		
	No	23 (14.5%)	26 (8.2%)	0.5 (0.29-0.9)			
Measles Vaccination	Yes	141 (90%)	259 (81.5%)	1	0.004		
	No	18 (11%)	59 (18.6%)	2.17 (1.2-3.6)			
Method of drawing of water from storage container	Pouring	Yes	58 (36.5%)	93 (29.2%)	2.07 (1.1-3.8)	0.019	
		No	101 (63.5%)	225 (70.8%)	1		
	Dipping	Yes	58 (38.4%)	101 (29.2%)	0.72 (0.4-1.0)	0.11	
		No	93 (61.6%)	225 (70.8%)	1		
Mother / caretaker diarrhea history	Yes	14 (8.8%)	13 (4.1%)	1	0.040		
	No	145 (91.2%)	305 (95.9%)	0.4 (0.2-0.96)			
Mother / caretaker finger nail cutting	Yes	154 (96.9%)	296 (93.1%)	1	0.101		
	No	5 (3.1%)	22 (6.9%)	2.28 (0.8-6.1)			
Finger nail cutting for child	Yes	153 (96.2%)	296 (93.1%)	1	0.175		
	No	6 (3.8%)	22 (6.9%)	1.8 (0.75-4.7)			
Hand washing material	Soap & water	77 (48.8%)	174 (69.3%)	1	0.075		
	Only water	73 (45.9%)	103 (31.2%)	0.6 (0.4-1.0)			
	Ash & water	9 (5.7%)	29 (9.1%)	1.4 (0.64-3.1)			
Methods of child feeding	Cup & spoon	Yes	88 (55.3%)	172 (54.1%)	1	0.79	
		No	71 (44.7)	146 (45.9%)	1.05 (0.7-1.5)		
Hand washing Time	Before food preparation & eating	Yes	131 (82.4%)	256 (80.5%)	1	0.620	
		No	28 (17.6%)	62 (19.5%)	1.13 (0.6-1.8)		
	After eating	Yes	40 (25.2%)	55 (17.3%)	0.62 (0.3-0.9)		0.044
		No	119 (74.8%)	263 (82.7%)	1		
Hand washing Time	After eating	Yes	135 (85%)	271 (85.2%)	1	0.928	
		No	24 (15%)	47 (14.8%)	0.9 (0.57-1.6)		
	After visiting latrine	Yes	71 (44.7%)	152 (68.2%)	1		0.517
		No	88 (34.6%)	166 (52.2%)	0.8 (0.60-1.2)		
After cleaning of child bottom	Yes	97 (61%)	214 (65.3%)	1	0.175		
	No	62 (39%)	104 (32.7%)	0.76 (0.5-1.1)			
		Yes	70 (44%)	166 (52.2%)	1	0.93	
		No	89 (56%)	152 (47.8%)	0.72 (0.4-1.0)		

In Bivariate analysis Age of child, family income, Latrine availability, Daily water consumption, Bottle feeding, Separate can take drinking water from storage, Method of drawing of water from the storage container (pouring), Mother / caretaker diarrhea history supplementary feeding commenced time, Measles vaccination were nominated with p-value  $\leq 0.25$  for multivariable logistic regression of under-five age childhood diarrhea morbidity.

### Determinants of Child Hood Diarrhea Morbidity

The Odds of childhood diarrhea morbidity among children who started supplementary feeding before six months was about five times higher when compared with their counterpart [AOR=5.38 95% CI (3.23-8.94)]. Similarly, the Odds of childhood diarrhea morbidity among children who started supplementary feeding after six months was about six times higher when compared with those started on 6 months. [AOR= 6.19 95% CI (2.96-12.93)] Table 5.

The Odds of childhood diarrhea morbidity among children who have no Measles vaccination was higher when compared with those who receive Measles vaccination with odds of [AOR=3.72 95% CI (1.609-8.63)].

**Table 5. Multivariate Analysis of Factors Associated with Children Under 5 Years of Age Diarrhea morbidity In Nekemte Town, Western Ethiopia**

Variables	Category	Diarrhea status		AOR (95% CI)
		Cases (%)	Control (%)	
Monthly income	Less than 500	51 (32.1%)	57 (17.9%)	1
	501-1000	27 (17%)	66 (20.8%)	0.863 (0.491-1.51)
	>1000	81 (50.9%)	195 (61.3%)	0.435 (0.263-0.72)
Supplementary feeding commenced time	On 6 months	62 (39%)	189 (59.4%)	1
	Before 6 months	78 (49.1%)	43 (13.5%)	5.38 (3.23-8.94)
Measles vaccination	After 6 months	15 (9.4%)	53 (16.7%)	6.19 (2.96-12.93)
	Yes	141 (90%)	259 (81.5%)	1
	No	18 (11%)	59 (18.6%)	3.72 (1.6-8.63)

### DISCUSSION

The major predictors of diarrhea morbidity in Nekemte town were family income, Supplementary feeding commenced time and Measles vaccination.

According to this study factors remained independently significant to the risk of diarrhea, were family income [AOR: 0.435, 95% CI: (0.263-0.72)], Supplementary feeding commenced time [AOR: 5.38, 95% CI: (3.23-8.94)] and Measles vaccination [AOR: 3.72, 95% CI: (1.6.9-8.63)].

From all socio-demographic factors tested only family income remained, significant after controlling other variables. This findings in line with other study conducted in Northern Ethiopia and Enderta Woreda, Tigray [22, 24].

From behavioral factors only supplementary feeding commenced time and measles vaccination were significant after controlling others variables. It was found that there was significant association between starting supplementary food lately after 6 months and childhood diarrhea morbidity which constitute with the study conducted in Haramiya and Kenya [23, 25].

The reason is that breast feeding and starting supplementary feeding on 6<sup>th</sup> months provides protective factors that may help reduce infections such as diarrhea, malnutrition and other infection for under five children. In addition, initiations of complementary feeding on 6 months may strengthened the immunity of children which indirectly reduces diarrhea causative organism accidentally introduced into supplementary foods during feeding practices and due to unhygienic procedures in the preparation of foods, materials and types of water used.

In this study, Children who were receive the measles vaccine had a lower risk of childhood diarrhea morbidity than those who did not receive measles vaccine with odds of 3.72 (1.609-8.63). This study is in line with the study conducted in India, in which Children who were given the measles vaccine had

a lower risk of diarrhea than their counterparts who had not been given the vaccine. Compared to unvaccinated children, measles vaccination was associated in reducing diarrhea in vaccinated children by 22% Democratic Republic of Congo, 12% in India, 21%t in Nigeria and 19.5 in Pakistan [26].

### **Limitation of the Study**

The limitation of this study was recalling bias, interviewer bias. It was difficult to measure some variable like income.

## **CONCLUSION**

In this study factors namely family income, Supplementary feeding commenced time and measles vaccination status were determinants of diarrhea morbidity among under five of age visiting in Nekemte town public health facilities. Thus, Nekemte town health office should aware mother/caregiver on supplementary feeding commenced time and measles vaccination. Further studies to identify the possible factor of childhood diarrhea.

## **ACRONYMY/ABBREVIATION**

AIDS	Acquired Immune Deficiency Syndrome
ARI	Acute Respiratory Infection
CI	Confidence Interval
DHS	Demographic Health Survey
EDHS	Ethiopian Demographic Health Survey
FMOH	Federal Ministry of Health
HC	Health Center
HEW	Health Extension Worker
HIV	Human Immune Deficiency Virus
NDDI	National Digestive Diseases Information Clearing house
ORS	Oral Rehydration Solution
UNICEF	United Nations Children's Fund
WHO	World Health Organization

## **DECLARATIONS**

### **Ethics Approval and Consent to Participate**

The research project was reviewed by an Institutional Review Board of Jimma University. Permission to conduct the research was obtained from the authorities in the study settings and written informed consents were secured from each participant.

### **Availability of Data and Materials**

Datasets used and /or analyzed during the current study available from the corresponding author on reasonable request.

### **Competing Interests**

The authors declare that they have no competing interests.

### **Authors' Contributions**

MG, TT, EA and YG participated from the inception of the research idea to proposal development, data collection, analysis and preparation & revision of the manuscript for publication. The authors read and approved the final version of the manuscript.

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## APPENDIX

## Questionnaires (English Version) Study Participants

1. Cases
2. Controls

Name of Health facility \_\_\_\_\_ Code number of the child (patient) -----

Name and signature of data collector: \_\_\_\_\_

Code of questions	QUESTIONS	Responses	Skip to
<b>PART I. SOCIOECONOMIC AND DEMOGRAPHIC CHARACTERISTICS</b>			
101	family's dwelling areas	1. Urban 2. Rural	
102	Relation of the respondent to the child	1. Mother 2. Caretaker	
103	Age of the mother/caretaker	Years 1. Currently Married 2. Divorced	
104	Marital status of the mother/caretaker	3. Single 4. Widowed 5. Separated	
105	Family size of the household	1. < 3 2. 3-5 3. 6-8 4. 9-11 5. >=12	
106	Number of U5 children	1. 1 2. 2 3. 3 4. >3	
107	Ethnic group of parents/caretakers	1. Oromo 2. Amhara 3. Others (Specify):	
108	Educational level of mother/caretaker	1. Formal education (A. Primary, B. Secondary, C. Higher) 2. Read and write 3. Read only 4. Neither write nor read	
109	Occupation of mother/caretaker	1. House wife 2. Government employee 3. Merchant 4. Farmer 5. Other (specify):	
110	Age of the child's father	Years 1. Formal education (last grade completed) 2. Read and write 3. Read only 4. Neither	
111	Educational level of the father	1. Government employee 2. Merchant 3. Farmer 4. No job 5. Other (specify)	
112	Occupation of the father	Muslim Orthodox Christian Others (specify): _____	
113	Religion	1. < 500 birr 2. 501-1000 birr 3. > 1000 birr 4. Do not know (unspecified)	
114	Monthly income		
<b>PART II. HOUSEHOLD ENVIRONMENTAL HEALTH CONDITIONS</b>			
201	Do have latrine facility?	1. Yes 2. No	205
202	Ownership of the latrine	1. Privately owned 2. Shared with neighbors	
203	Type of latrine facility (observation)	1. Traditional pit latrine 2. VIP latrine 3. Public latrine	

Code of questions	QUESTIONS	Responses	Skip to
204	Does the latrine currently functioning? If the family has no latrine, where do you dispose	4. Communal latrine 5. Other (specify): 1. Yes 2. No	
205	human waste (Adult member open defecation)	1. Open field 2. Other (specify)	
206	How do you dispose refuse? (Multiple response is possible)	A. Pit B. Open field C. Burning D. Garbage can E. Other (specify):	
207	Is feces seen around the house (or in the compound)?	1. Yes 2. No	
208	Where you dispose infant feces/excreta?	1. Not in latrine 2. In latrine	
209	Status of drinking water source	1. Improved 2. Unimproved	
210	Daily water consumption (Per capita water consumption)	1. ≤ 20 Liters 2. > 20 Liters	
211	Distance from the house to the water source	Minutes	
212	Number of rooms in the house	1. 1 2. 2 3. 3 and above	
213	Are there domestic animals living in the same house with the members of the family?	1. Yes 2. No	
<b>PART III: BEHAVIORAL ASPECTS</b>			
301	Did you wash your hands yesterday?	1. Yes 2. No	→ 304
302	At what point/time?	1. Before food preparation and eating 2. After eating 3. After visiting latrine 4. After cleaning of child bottom 5. Other (specify):	
303	By using what did you wash hands yesterday?	1. Soap & water 2. Ash & water 3. Only water 4. Other (specify):	
304	Do you separately prepare food for the child, using a Separate material?	1. Yes 2. No	
305	Do you cut your child nail when grown?	1. Always 2. Sometimes 3. Never	
306	What do you use to feed the child?	1. Hand 2. Cup and spoon 3. Cup 4. Bottle 5. Other (specify):	
307	Does the drinking-water storage container have a cover? Ask the respondent to show you the storage container	1. Yes 2. No	
308	Is there a separate can for taking drinking water from the storage container? Ask the respondent to show you the Can	1. Yes 2. No	
309	Type of collection container	1. Pot 2. Plastic bucket 3. Iron bucket 4. Jerry can 5. Other (specify):	
310	Method of drawing of water from the storage container		
	Dipping	1. Yes 2. No	
	Pouring	1. Yes	

Code of questions	QUESTIONS	Responses	Skip to
		2. No	
311	Others (specify).... Covering material is used during transportation	1. Yes 2. No	
312	Frequency of washing collection containers and others per week	1. Not washed 2. Once 3. Twice 4. $\geq$ Three	
	When is strict care while handling water?		
	Fetching	1. Yes 2. No	
313	Transporting	1. Yes 2. No	
	Storing	1. Yes 2. No	
	Drinking	1. Yes 2. No	
314	Others Do you treat your water in any way to make it safer to drink?	1. Yes 2. No	
315	What do you usually do to the water to make it safer to drink?	1. Boil 2. Chlorine 3. Strain through cloth (Filtering) 4. Other (specify): _____	
<b>PART IV: INFORMATION OF THE INDEX CHILD</b>			
401	Age of the index child (in Months)	1. 0-5 Months 2. 6-11 3. 12-23 4. 24-35 5. 36-47 6. 48-59	
402	Sex of the index child	1. Male 2. Female	
403	Birth order of the child	1. First 2. Second 3. Third 4. Fourth & above	
404	Do you (the mother/caretaker) have a history of diarrhea in the past two weeks?	1. Yes 2. No	
405	Have you ever breast-fed your child?	1. Yes 2. No	407
406	For how long did you breastfed your child?	1. <1 Year 2. $\geq$ 1 Years	
407	What is his/her current breastfeeding status?	1. Exclusive breastfeeding 2. Partial breastfeeding 3. Not breastfeeding	
408	At what age the child started supplementary /weaning food?	1. Before 6 Months 2. At 6 Months 3. After 6 Months	
409	Did the child receive measles vaccination? Ask for	1. Yes (response of the respondent) 2. Yes, (by checking the card) 3. No	
410	children of age greater than nine months Do your child have diarrhea today? (Only for Cases)	1. Yes 2. No	End
411	For how long the diarrhea last?	1. Less than 14 days 2. Greater than 14 days	
412	If the child has diarrhea today, how many times a day he/she passes stool?	1. Three times 2. Less than three times 3. More than three times 4. Don't know	
413	The type of diarrhea that the child had	1. Watery 2. Blood and mucus	